# Inositol phosphate in the basidiomycete fungus *Schizophyllum commune*

**Dissertation**

To fulfill the Requirements for the Degree of
**„doctor rerum naturalium“ (Dr. rer. nat.)**

**Submitted to the Council of the Faculty**
**of Biological Science**
**of the Friedrich Schiller University Jena**

**by Reyna Carmina Felicia Murry, MSc.**
**born on the 22$^{nd}$ of October 1987 in Bandung, Indonesia**

First reviewer: Prof. Dr. Erika Kothe, Institut für Mikrobiologie, FSU Jena
Second Reviewer: Prof. Dr. Axel A. Brakhage, Institut für Mikrobiologie, FSU Jena
Third Reviewer: Prof. Dr. J. Stephen Horton, Department of Biological Sciences, Science and Engineering Center, Union College, Schenectady, New York, USA

Date of public defense: 7th May 2019

“*Science is the key to our future, and if you don't believe in science, then you're holding everybody back.”*

*Bill Nye*

# Table of Contents

1 Introduction ........ 1

1.1 *Schizophyllum commune* ........ 1

1.2 Fruiting body development in *S. commune* ........ 3

1.3 Signal transduction: G-protein-coupled receptors (GPCRs) and Ras signaling ........ 4

1.4 Inositol-based metabolism and signaling: inositol phosphates and inositol lipids ........ 5

1.5 Inositol and inositol monophosphatase ........ 7

1.6 Inositol hexakisphosphate and the inositol phosphate kinases ........ 8

1.7 Aim of the study ........ 8

2 Materials and methods ........ 10

2.1 Chemicals, reagents and enzymes ........ 10

2.2 Cultivation media ........ 10

2.3 Organisms, plasmids, and nucleotides ........ 11

2.4 Experiments with *E. coli* DH5α ........ 13

2.4.1 Growth conditions and production of electrocompetent cells ........ 13

2.4.2 Transformation of *E. coli* DH5α ........ 13

2.5 Experiments with *S. commune* ........ 14

2.5.1 Cultivation of *S. commune* ........ 14

2.5.2 Growth rate measurement and dry weight determination ........ 14

2.5.3 Rapid DNA isolation ........ 14

2.5.4 DNA isolation using CTAB ........ 15

2.5.5 RNA isolation ........ 15

2.5.6 Quality, purity, and quantitation for DNA and RNA ........ 16

2.6 Inositol monophosphatase activity measurement ........ 16

2.6.1 Crude extraction of *S. commune* ........ 16

2.6.2 Protein quantitation ........ 16

2.6.3 Malachite Green assay ........ 17

2.7 Protoplast preparation and transformation of *S. commune* an ........ 17

2.8 Construction plasmid: *imp* overexpression for *S. commune* ..... 18
  2.8.1 Yeast Recombination ..... 18
  2.8.2 Cultivation of *S. cerevisiae* BY4741, Δ*LEU* ..... 19
  2.8.3 Yeast transformation ..... 20
  2.8.4 Screening and validation of recombined product ..... 20
  2.8.5 Cloning of *tef1* promoter and *imp* gene to pTrp ..... 21
2.9 Proteome analysis using 2D-PAGE ..... 21
  2.9.1 Cytosolic protein isolation ..... 21
  2.9.2 First and second dimension of 2D gel electrophoresis ..... 22
  2.9.3 Protein/ peptide analysis using MALDI TOF/MS-MS ..... 23
2.10 Signal protein analysis ..... 24
  2.10.1 Signal protein isolation and purification ..... 24
  2.10.2 LC-MS/MS analysis and protein database search ..... 25
2.11 Microscopy ..... 26
  2.11.1 Stereomicroscopy and fluorescence microscopy ..... 26
  2.11.2 Confocal Laser Scanning Microscopy ..... 26
2.12 Inositol phosphates profile analysis ..... 27
  2.12.1 Radioactive labelling and extraction of inositol phosphates ..... 27
  2.12.2 HPLC run and detection using scintillation counter ..... 27
2.13 Other molecular biology methods ..... 28
  2.13.1 Polymerase chain reaction (PCR) ..... 28
  2.13.2 Real Time-quantitative PCR (RT-qPCR) ..... 29
  2.13.3 DNA restriction endonucleases ..... 29
  2.13.4 DNA sequencing ..... 29
  2.13.5 Plasmid DNA preparation ..... 30
  2.13.6 Determination of Protein Kinase C activity ..... 30
2.14 *In silico* analysis ..... 30
  2.14.1 Inferring phylogeny ..... 30
  2.14.2 Chromosomal organization of *imp* surrounding gene ..... 31

2.14.3 Other protein analysis 31

3 Results 32

3.1 Inositol phosphate in *S. commune* 32

3.2 Inositol phosphate phosphatase and inositol phosphate kinases 34

3.3 Inositol phosphate kinases gene expression 36

3.4 Proteins involved in the development of *S. commune* 37

3.5 Lithium effect on inositol phosphate in *S. commune* 39

3.5.1 Lithium effect on the growth and mating in *S. commune* 39

3.5.2 Lithium effect on inositol phosphate and inositol monophosphatase activity 41

3.5.3 Lithium effect on *S. commune* metabolism and cellular signaling 42

3.5.4 Lithium effect on actin dynamics 44

3.6 Inositol monophosphatase and inositol monophosphatase-like gene family 45

3.7 Inositol monophosphatase 46

3.8 Comparative genetic map of *imp* and surrounding genes 48

3.9 Overexpression of inositol monophosphatase (*imp*) 50

3.9.1 Validation of *imp* overexpression 50

3.9.2 Characterization of *imp* overexpressing mutants 51

3.9.3 Mating interaction of *imp* overexpressing mutants 53

3.9.4 Inositol phosphates profile in *imp* overexpressing mutants 54

3.9.5 Effect of SDS on *imp* overexpressing mutants 55

3.10 Proteome analysis in *imp* overexpressing mutants 56

3.11 Membrane trafficking related proteins are induced in *imp* overexpressing strains 58

3.12 The effect of Brefeldin A on membrane trafficking in *imp* overexpressing mutants 61

3.13 Inositol monophosphatase in developmental stages of *S. commune* 63

3.14 Putative crosstalk between Ras1 and Imp 64

3.14.1 Ras1 activation alters inositol phosphates profile in the presence of lithium 65

3.14.2 Gene expression of *ras1* and *ras2* prior to *imp* overexpression 67

3.14.3 Mating and fruiting bodies of Ras1 active mutants and *imp* overexpressing mutants 68

4 Discussion ... 70
  4.1 Inositol as a building block for inositol phosphate and phosphoinositide ... 70
    4.1.1 Inositol phosphate metabolism in *S. commune* ... 71
    4.1.2 Inositol polyphosphates play role in sexual development of *S. commune* ... 72
  4.2 Lithium effect on inositol phosphate signaling ... 73
    4.2.1 Lithium attenuates inositol monophosphatase and alters inositol phosphates ... 73
    4.2.2 Lithium affects fungal growth and sexual development ... 75
  4.3 The role of Inositol monophosphatase ... 77
    4.3.1 Inositol monophosphatase and inositol generation ... 77
    4.3.2 Inositol monophosphatase is important for intracellular trafficking ... 79
    4.3.3 Potential role of inositol monophosphatase in cell wall integrity (CWI) ... 80
  4.4 Ras signaling is interrelated with inositol phosphate signaling ... 81
    4.4.1 Possible crosstalk between Ras and inositol signaling ... 81
    4.4.2 Lithium action in Ras and inositol phosphate signaling ... 82
  4.5 Sexual development of *S. commune* ... 83
5 Outlook ... 89
6 Summary ... 91
7 Zusammenfassung ... 93
8 References ... 95
9 Supplemental Material ... 105
10 Abbreviations ... 286
Statement of authorship ... 288
Curriculum vitae ... 289
Acknowledgement ... 291

# 1 Introduction

## 1.1 *Schizophyllum commune*

*S. commune* is known as a split-gill mushroom and a white rot fungus (Figure 1). This mushroom-forming fungus is globally distributed all over continents except Antarctica. This fungus naturally lives saprophytically on dead woods (Figure 1 A-B). *S. commune* produces extracellular ligninolytic enzymes which degrade the complex organic polymer lignin and causes woods to decay (Asgher *et al.*, 2016; Ganeshpurkar *et al.*, 2010), leaving behind the lighter-whitish-yellowish-color on a wood surface. *S. commune* has recently been acknowledged for its medical value since it produces the biopolymer schizophyllan that possesses antitumor activity (Okazaki *et al.*, 1995). Further, schizophyllan is also utilized in polymer-based oil recovery processes (Gao, 2016).

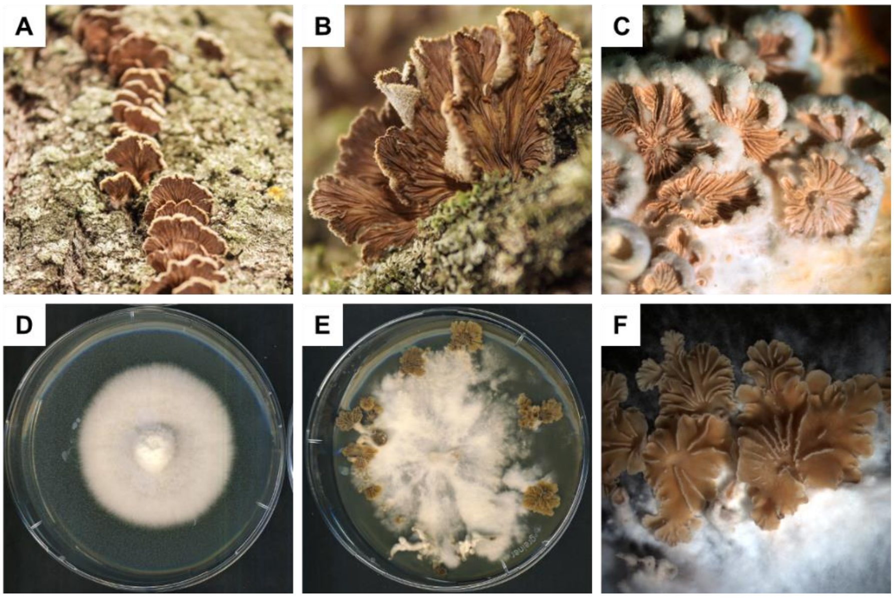


Figure 1. The wood-inhabiting fungus *S. commune* on its natural habitats beech timber and dead logs found in Paradies park, Jena, Germany (A, B). *S. commune* is cultivated under laboratory conditions on artificial sorghum-branch (C) and on minimal media plate (D, E, F). Monokaryon fungus shows a regular form of mycelia (D) and dikaryon strain shows irregular mycelia and producing fertile fruiting bodies on petri dish plate (E, F).

*S. commune* can be simply cultivated using synthetic media under laboratory conditions (Figure 1D-F) and it is able to complete its life cycle within 10-14 days. The complete fully sequenced genome of *S. commune* strain H-48 was published in 2010. *S. commune* had been selected as a model basidiomycete due to the fact that genetic manipulation is possible to perform (Munoz-Rivas *et al.*, 1986; van Peer *et al.*, 2009). The techniques that can be applied to study *S. commune* are RNA-mediated gene silencing (de Jong *et al.*, 2006), gene deletion by homologous recombination (Ohm *et al.*, 2010a; Schubert *et al.*, 2006), and gene over-expression (Kirtzel *et al.*, 2018; Yamagishi *et al.*, 2005). *S. commune* has been featured as a model organism to study mating type genes (Freihorst, 2015; Freihorst *et al.*, 2016; Kothe, 1999; Raper and Raper, 1966), mushroom development (Ohm *et al.*, 2013; Ohm *et al.*, 2011), ligninolytic enzymes (Hirai *et al.*, 2008; Madhavan *et al.*, 2014), and GPCRs signaling in response to pheromone regulation (Erdmann *et al.*, 2012; Kothe *et al.*, 2009; Raudaskoski and Kothe, 2010).

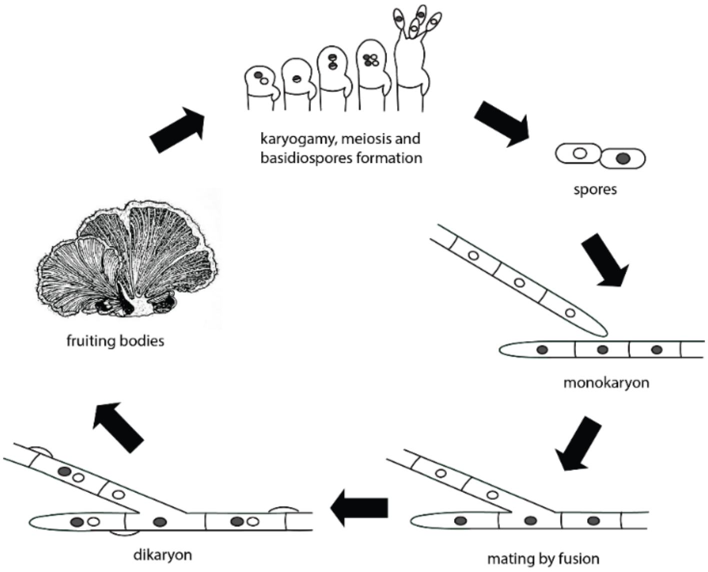


Figure 2. The life cycle of *S. commune*. Basidiospores are formed at basidia inside a fruiting body. Mature basidiospores germinate and develop to monokaryotic mycelium. Fertile dikaryon only occurs if two monokaryons with compatible *matA* and *matB* mating types fuse. The mating type locus that controls each of steps in this process is described in this figure. Formation of fruiting bodies requires suitable conditions, such as low carbon dioxide intensity and sufficient light.

Life cycle and sexual reproduction in the basidiomycete *S. commune* are typical of the group of gilled mushrooms (Raper and Miles, 1958). This fungus is classified as heterothallic, which means the fusion is only possible between two haploids from different alleles at the mating type locus. During sexual reproduction, two haploid monokaryotic hyphae of opposite mating types encounter. Hyphal fusion of two monokaryotic hyphae (so-called anastomosis) triggers signaling compounds exchange, such as lipopeptide pheromone that differentiate each other's mating type and triggers anastomoses. This event is controlled by the tetrapolar mating system comprising two independent genetic factors *A* and *B* mating type loci (Figure 2). The *A* genes are positioned at scaffold 1 and the *B* genes at scaffold 10 (Ásgeirsdóttir *et al.*, 1994; Ohm *et al.*, 2010b). The *A* genes (homeodomain genes) of different allelic specificities interact with each other through a formation of heterodimers of homeodomain proteins which play a role as transcription factors. The *B* genes (pheromone and receptor genes) encode pheromones and pheromone receptors. Pheromone receptors as a part of GPCRs activate downstream signaling cascade such as Mitogen-Activated Protein-Kinase (MAP-K) (Raudaskoski and Kothe, 2010). Dikaryotization involves series of events including nuclear migration, nuclear pairing & clamp cell formation, conjugated division, septa formation and hook cell fusion, which are factor *A* or *B* dependent (Figure 2). These serial events take place in a basidiomycete-specific structure called clamp cell. Mature fruiting body bearing haploid basidiospores is formed and basidiospores will be germinated into monokaryotic mycelium.

## 1.2 Fruiting body development in *S. commune*

Fruiting body development commences with the aggregation of aerial dikaryotic mycelia. The mycelial aggregate from dikaryotic mycelia, known as primordia (Figure 3B, E) is described as a small bump or knob-like structure on the surface of aerial mycelia. Later, the primordia develop to mature fruiting bodies (Figure 3C, F). In the basidiomycete, an appropriate environmental condition is required to form and to develop mature fruiting bodies. Light, carbon dioxide, temperature, humidity, pH, nutrients availability including source of nitrogen and carbon are the crucial factors that can influence the fruiting body formation (Bellettini *et al.*, 2016; Ohm *et al.*, 2010b; Palmer and Horton, 2006; Wessels, 1993).

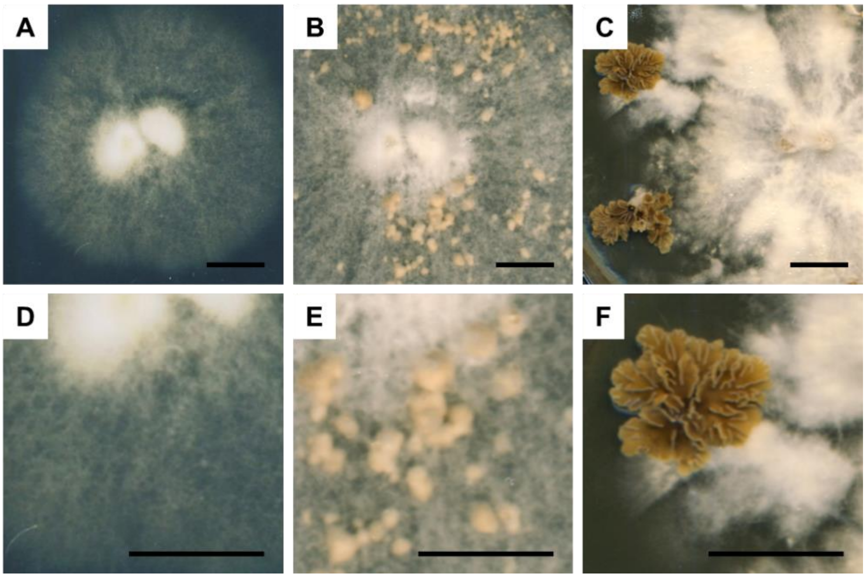


Figure 3. Mushroom development of *S. commune* from monokaryotic mycelia to the fruiting body on MM (A-F). Aerial hyphae of dikaryotic strains (A, D) develop further to knob-like structure so-called primordia (B, E). The primordia later form basidiospores-bearing fruiting bodies.

Light is one of the most important aspects of mushroom development in basidiomycete fungi including *S. commune* (Eger-Hummel, 1980; Robert and Durand, 1979). A short exposure of light is allowed to induce primordia (Perkins, 1969) and intensive light exposure is required for further stages of fruiting body development. Blue light is known to initiate fruiting body formation in *S. commune,* which is detected by the white collar (WC) complex comprising of White Collar 1 (WC-1) and White Collar 2 (WC-2). *S. commune* possesses both WC-1 and WC-2, which are predicted as light receptor and as a transcriptional regulator, respectively (Ohm *et al.*, 2013). Several transcription factors such as *hom1*, *hom2*,*fst3*,*fst4, bri1*, *c2h2*, and, *gat1* play a role in fruiting body formation of *S. commune.* Gene *bri1* and *hom2* are involved in the formation of aggregates from dikaryotic aerial hyphae. Gene *fst4* and *c2h2* are required in primordia formation. Gene *fst3* acts as a repressor for primordia formation. On the other hand, *hom1* and *gat1* are essential for tissue development in *S. commune* (Ohm *et al.*, 2011).

Hydrophobins specific for fruiting body, lectin, haemolysins, expansins, and several oxidative enzymes are group of proteins that are essential in fruiting body development (Boulianne *et al.*, 2000; Bouzarelou *et al.*, 2008; Madhavan *et al.*, 2014; Muraguchi and Kamada, 2000; Muraguchi *et al.*, 2015; Vidic *et al.*, 2005; Wessels *et al.*, 1991). Hydrophobins are retained in the aggregation of aerial hyphae and formation of fruiting bodies (Wessels *et al.*, 1991). In *Coprinopsis cinerea*, carbohydrate binding related proteins so-called lectins *cgl1* and *cgl2* are differentially regulated during development of fruiting bodies. The expression of *cgl2* is represented in the early stages of fruiting body formation especially in the hyphal knot formation and maintained until maturation of the fruiting body, whereas *cgl1* is particularly expressed in primordia and mature fruiting bodies (Boulianne *et al.*, 2000). Oxidative enzymes such as laccases and multicopper oxidases (MCOs) were found to be associated with the fungal morphogenesis in *S. commune* (Madhavan *et al.*, 2014). Cytochrome P450 is presumably involved in development process of mushroom. In *Agaricus bisporus* and *Antrodia cinnamomea*, cytochrome P450 is exclusively expressed only in fruiting body and inactivated in mycelial stage (Hsu *et al.*, 2011). Whereas, in *Coprinus cinereus*, cythocrome P450 is associated with formation of primordial short stipes until mature fruiting bodies are established (Muraguchi and Kamada, 2000). Expansin has a function in loosening cell wall and cell wall extension and facilitates growth of cell walls and disturbs the non-covalent linkages that retain microfibrils in the cell wall. (Cosgrove, 2005; Sampedro and Cosgrove, 2005). Expansin was found in *S. commune* (Tovar-Herrera *et al.*, 2015) and presumably involved in cell wall enlargement during mushroom development. Fungal hemolysins are pore-forming proteins that form aggregation on the cell surface after the detection of specific membrane components (Nayak *et al.*, 2013). Ostreolysin and aegerolysin are examples of fungal hemolysin which are found in *Pleurotus ostreatus* and *Agrocybe aegerita*. Both lysins are important for fruitification (Berne *et al.*, 2002) and they were found particularly in the promptly growing primordia, basidia and basidiospores (Vidic *et al.*, 2005).

## 1.3 Signal transduction: G-protein-coupled receptors (GPCRs) and Ras signaling

Heterotrimeric GPCRs are the largest superfamily of integral membrane proteins conserved in fungi (Xue *et al.*, 2008) and they play roles in many cellular activities (Figure 4). They consist of seven transmembrane α-helical regions that are activated by a wide-spectrum of ligands (Kobilka, 2007). After activation by one of extracellular ligand, the intracellular GPCR changes its conformation and subsequently activate G-proteins. This event triggers the exchange of guanosine diphosphate (GDP)- guanosine trisphosphate (GTP) associated with the Gα subunit and dissociation between Gβ/Gγ dimer and Gα subunit occurs. The disassociated Gβ/Gγ dimer and Gα remain attached to the plasma membrane, but they are no longer bound to GPCR. However, they become free to interact with their downstream effectors/transducer and they deliver the signal to different second messengers (Tuteja, 2009). The effector/transducer is responsible for sending the signal from first to second messenger, which could be regulated by intracellular protein kinases or phosphatases. The possible targets of the signal are enzymes, intracellular receptors, special transport vehicles and transcription factors that control the gene expression. The possible second messengers are as following: cyclic-adenosine monophosphate (cAMP) which is activated by adenyl cyclase, cyclic-guanosine monophosphate (cGMP) which is activated by guanylate cyclase, inositol-1,4,5-triphosphate ($IP_3$) and diacyl glycerol (DAG) which are generated by phospholipase-C (PLC) upon cleavage of phosphatidylinositol bisphosphate ($PIP_2$) (Berridge, 1984), calcium ion ($Ca^{2+}$) whose its activation is triggered by $IP_3$ receptor (Kiselyov *et al.*, 2003), and Ras GTPase upon activation by Ras guanine exchange factors (GEFs). In fungi, GPCRs signaling is involved in many crucial developmental processes such as growth, mating and pathogenesis (Bardwell, 2005; Bolker, 1998; Li *et al.*, 2007; Servin *et al.*, 2012).

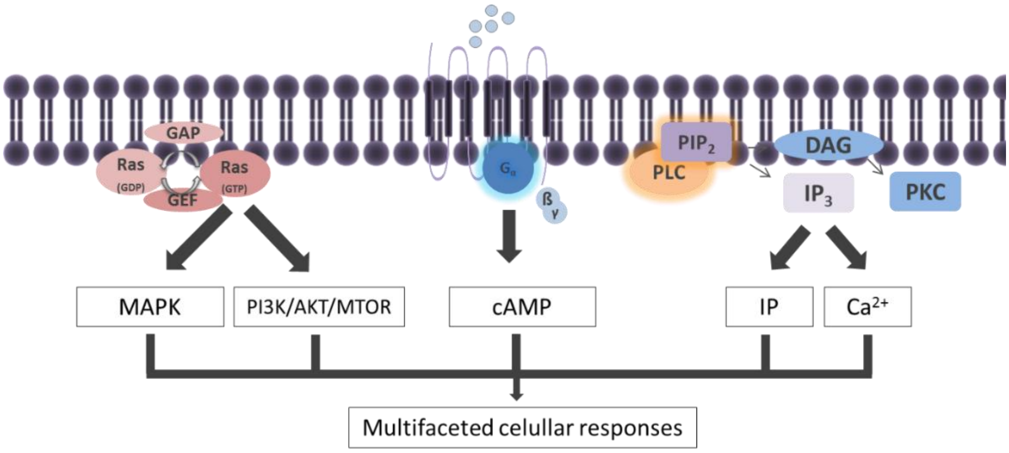


Figure 4. G-coupled protein receptors (GPCRs) and Ras signaling control a multifaceted cellular signaling. MAP-Kinase, PI3K/AKT/MTOR, cAMP, and Phosphatidylinositol (PI) and Inositol phosphate signaling are the downstream signaling of GPCRs and Ras signaling.

In *S. commune*, the small GTPase protein Ras are classified into five subfamilies: Ras, Rho, Rab, Ran and Arf. (Raudaskoski *et al.*, 2012). The activation of small GTPase Ras is controlled with GEF (guanine nucleotide exchange factors). GEF promotes the exchange of GDP to GTP and allows the interaction of the GTP-bound GTPase with the downstream effectors. On the opposite, GTPase-activating proteins (GAPs) deactivate the G-protein Ras by recruiting the intrinsic GTPase activity. *S. commune* possesses two Ras protein, Ras1 and Ras2. Ras1 was extensively studied and known to play a versatile role in fungal growth, hyphal polarization, pheromone signaling, and sexual development. On the other hand, the role of Ras2 is still unknown. Furthermore, inositol signaling was found to be regulated through Ras signaling (Knabe *et al.*, 2013).

## 1.4 Inositol-based metabolism and signaling: inositol phosphates and inositol lipids

Inositol is a simple polyol that acts as a molecular building block for inositol phosphate and inositol lipids. Inositol possesses six hydroxyl (-OH) groups comprises one axial 2-hydroxyls and five equatorial hydroxyls (Irvine and Schell, 2001). Each can be substituted by phosphate group. Inositol phosphate phosphatases facilitate phosphorylation or addition of phosphate group to inositol and in contrast, inositol phosphate kinases work on dephosphorylation of inositol phosphates. Inositol phosphates are derived from the lipid inositol phosphatidylinositol 4,5 bisphosphate/ $PIP_2$ (Figure 5), a main inositol lipid in the Phosphatidylinositol signaling (Irvine, 2016).

The PI signaling system is present in all eukaryotic cells including fungi, however it is most intensively studied in mammals (Majerus *et al.*, 1999). A membrane-associated enzyme phosphoinositide-specific phospholipase C (PI-PLC) cleaves $PIP_2$ into two important second messenger inositol 1,4,5 triphosphate ($IP_3$) and diacylglycerol (DAG), involved in calcium release from intracellular stores and activation of protein kinase C (PKC) (Berridge, 1993; Hughes and Putney, 1990). $IP_3$ is either dephosphorylated by an inositol phosphate phosphatase to inositol 1,4-bisphosphate for re-generation of inositol, or alternatively, it is phosphorylated to inositol 1,3,4,5-tetrakisphosphate by inositol phosphate kinase (Hughes and Putney, 1990). The full phosphorylated inositol phosphates $IP_6$ can be further phosphorylated to inositol pyrophosphates ($IP_7$ and $IP_8$). Inositol pyrophosphates possess a diphosphate on one or more carbons of the inositol ring. The inositol pyrophosphates are high energy signaling molecules involved in various cellular processes, including regulation of cell death and telomere length (Saiardi *et al.*, 2005), modulation of vesicular trafficking (Saiardi *et al.*, 2002), regulation on chemotaxis and cellular development of *Dictyostelium discoideum* (Pisani *et al.*, 2014), and metabolic adaptation for host virulence on cryptococcal diseases (Lev *et al.*, 2015).

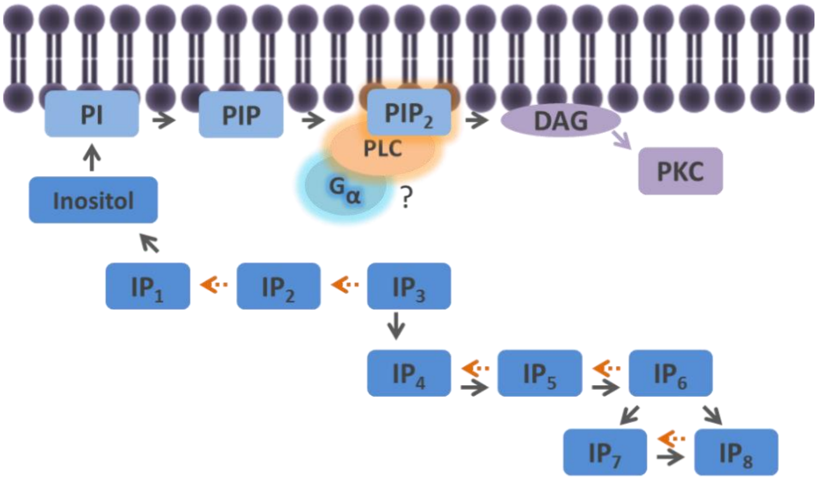


Figure 5. Inositol phosphate (IP) signaling pathway. The binding of extracellular stimuli to G-alpha subunit activates subsequently phospholipase C (PLC). PLC cleaves phosphatidylinositol 4,5 bisphosphate ($PIP_2$) generating two second messengers, inositol 1,4,5 triphosphate ($IP_3$) and diacylglycerol (DAG). The lithium sensitive inositol monophosphatase generates inositol from inositol monophosphate (IP1). Inositol is used to build up phosphoinositide using the phosphatidylinositol synthase (PI Synthase), and other inositol phosphates using inositol phosphate kinases. Abbreviations: Gα: G-alpha subunit; PI: phosphatidylinositol; PIP: phosphatidylinositol 5 phosphate; $PIP_2$: phosphatidylinositol 4,5 bisphosphate; DAG: diacylglycerol; PKC: protein kinase C; PLC: phospholipase C; $IP_1$: inositol monophosphate; $IP_2$: inositol bisphosphate; $IP_3$: inositol trisphosphate; $IP_4$: inositol tetrakisphosphate; $IP_5$: inositol pentakisphosphate; $IP_6$: inositol hexakisphosphate; $IP_7$: 5-diphosphoinositol-pentakisphosphate; $IP_8$: 5-bis-diphosphoinositol-tetrakisphosphate.

## 1.5 Inositol and inositol monophosphatase

Inositol monophosphatase is an enzyme that plays a critical role in dephosphorylating inositol monophosphate ($IP_1$) releasing inositol that is recycled back to phosphatidylinositol (PI).

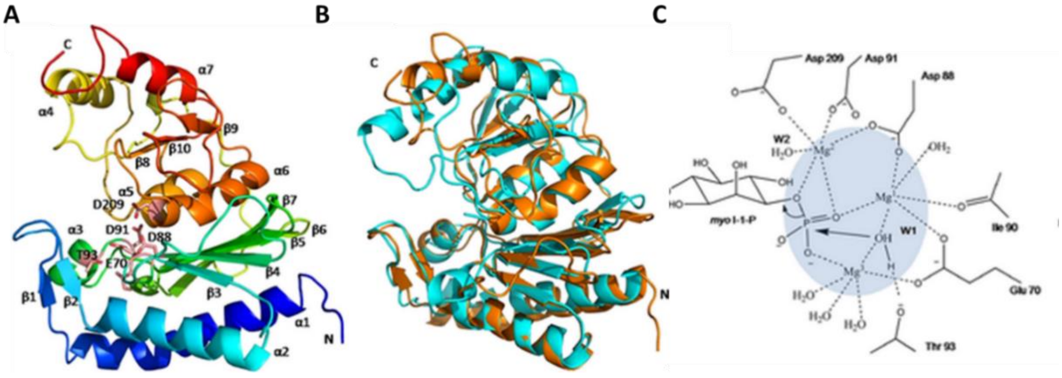


Figure 6. Three-dimensional crystallography structure of *Staphylococcus aureus* inositol monophosphatase (SaIMPase) consists of five alternative layers of alpha helices and beta sheets (αβαβα). Blue to red depicts N-terminal to C-terminal amino acids, respectively. Two domains indicate the active-site metal binding Glu70, Asp88, Asp91 and Asp209, together with Thr93 (pink residues), B. Alignment of SaIMPase (orange) and human inositol monophosphatase (blue) (Dutta *et al.*, 2014).

Inositol is a building block molecule for phosphoinositide or inositol phosphates, thus inositol monophosphatase is very important for the generation of membrane associated-phosphatidylinositol and cytosolic inositol phosphate signaling. The enzymatic function of Inositol monophosphatase and its structural properties are relatively well characterized in mammalian but not in fungi, however little is known about the regulation of the enzyme (Berggard *et al.*, 2002).

Inositol monophosphatase is uncompetitively inhibited by lithium and has been proposed as the putative target of the inositol depletion (Pollack *et al.*, 1994). Inositol monophosphatase gained popularity as the putative molecular site of action of lithium therapy for bipolar disease (Bone *et al.*, 1992). For its catalytic activity, inositol monophosphatase depends on three magnesium ions as cofactor (Lu *et al.*, 2012). Restricted analysis showed that certain region in inositol monophosphatase sequence share similarity with protein inositol polyphosphate 1-phosphatase (IPP) and fructose 1,6-biphosphatase (F1,6-BP), specifically in the region which is relevant to substrate-metal binding and nucleophile activation (Atack, 1995; Neuwald *et al.*, 1991). Each subunit of inositol monophosphatase presents as an alternating alpha-helices and beta-sheets secondary structures organized in a five-layered alpha-beta-alpha-beta-alpha sandwich (Figure 6A).

This unique structure is similarly found in F1,6-BP and IPP (Atack *et al.*, 1995). In mammals, lithium inhibits inositol monophosphatase activity in vitro, thus regulates phosphatidylinositol signaling at therapeutically relevant concentrations (0.5-1.0 mM) (Atack *et al.*, 1995). Similar pattern of enzyme inhibition in a higher concentration (100-200 mM) was found in model yeast *Saccharomyces cerevisiae* (Murray and Greenberg, 1997). Inositol monophosphatase is able to bind to calbindin, a member of the calmodulin superfamily of $Ca^{2+}$ binding proteins, thereby calbindin increases the phosphatase activity of inositol monophosphatase (Berggard *et al.*, 2002). Inositol monophosphatase has a broad spectrum of functional role interkingdom. In plants, inositol monophosphatase has a functional role in the histidine biosynthetic pathway and catalyzes the hydrolysis of inositol and galactose-phosphates (Nourbakhsh *et al.*, 2014). The regulation of plant *imp* is induced by cold stress (Zhang *et al.*, 2017). On the other hand, in euryhaline eel *Anguilla anguilla,* inositol monophosphatase controls osmoregulation and adaptation of eel in high salinity environment (Kalujnaia *et al.*, 2010).

## 1.6 Inositol hexakisphosphate and the inositol phosphate kinases

Inositol phosphates are a multifaceted group of second messengers generated by the serial phosphorylation of lipid derivative inositol1,4,5-trisphosphate ($IP_3$). Inositol hexakisphosphate is pinpointed as the most abundant inositol phosphate in eukaryote (Sasakawa *et al.*, 1995) and a plant major storage of phosphorous element (Raboy *et al.*, 2001). $IP_6$ is linked to various cellular process and exclusively known to be crucial for mRNA export from the nucleus in *S. cerevisiae* and human cells (York *et al.*, 1999). $IP_6$ stimulates the repair mechanism of double strand breaks by non-homologous DNA end joining pathway through $IP_6$–ku binding (Ma and Lieber, 2002) or $IP_6$–DNA-PKcs binding (Hanakahi *et al.*, 2000). $IP_6$ inhibits serine-threonine phosphatases, thus allowing activation of L-type calcium channel in pancreatic cells (Larsson *et al.*, 1997). $IP_6$ is accountable for endocytosis role by modulating dynamin I-mediated endocytosis in the human pancreatic β cell (Høy *et al.*, 2002) and by binding to a clathrin assembly protein AP3 (Norris *et al.*, 1995). Inositol phosphate multikinase (Imk) is the enzyme that responsible to catalyze the phosphorylation of the most prominent second messenger $IP_3$ to the full phosphorylated inositol phosphate hexakisphosphate ($IP_6$). Imkadditionally synthesizes the precursors of $IP_6$, namely $IP_4$ and $IP_5$. In addition to that, inositol pentakisphosphate 2-kinase (Ipk) catalyzes specifically the synthesis of $IP_6$ from inositol 1,3,4,5,6- tetrakisphosphate, which is found in mammals, plants, *Drosophila melanogaster*, *S. cerevisiae* (Kuo *et al.*, 2014; Verbsky *et al.*, 2005).

## 1.7 Aim of the study

Fruiting body differentiation is one of the most important events in the sexual development of basidiomycete fungi. The development of fruiting body has been hypothesized occupying a complex cellular signaling ranging from GPCRs, MAP-Kinase, cAMP and Ras signaling. However, we are still in the beginning phase to understand the connection between each signaling cascade in conjunction with fruiting body development.

Ras, a small GTP binding protein is however known to play a central role in the sexual development phase by controlling polar growth, branching, nuclear migration, fruiting body morphology, and spore production. Molecular switching between GDP (inactive) and GTP (active) Ras is a key module that allow us to comprehend the importance of Ras signaling in sexual development. Constitutive activation of Ras influences regulation of a broad-spectrum cellular processes and metabolism, emphasizing inositol phosphate metabolism and inositol based signaling as a central topic for this study.

Inositol controls a wide-range signaling molecules by managing phosphate groups to be attached to the six-carbon inositol backbone, thus creating a tremendous amount of different inositol phosphates. Additionally, phosphate with its ability to connect between inositol and diacylglycerol (DAG) through the phosphodiester bond, resulting in lipid inositol phosphates (termed as phosphoinositide) that broaden signaling potential of this molecules group.

There are two main purposes of this study. First, this study aims to investigate the relationship between Ras activation and inositol phosphate signaling in the context of development of *S. commune*, as a potential cross-talk between inositol monophosphatase and Ras1 was found previously. Second, this investigation represents an attempt to elucidate the biological function of inositol phosphate in *S. commune*.

To investigate the role of inositol phosphate signaling in *S. commune*, several techniques were performed: (1) *in silico* analysis of the gene and proteins involved in the inositol-based metabolism and signaling, (2) gene expression analysis of inositol monophosphatase and inositol phosphate kinases, (3) genetic manipulation of inositol monophosphatase using overexpression, (4) proteome analysis in different sexual developmental stages, and (5) inositol phosphate detection in different sexual developmental stages.

Altogether, this study provides an overview of inositol phosphates metabolism and its potential signaling that may contribute to the development of mushroom forming fungus *S. commune*.

# 2 Materials and methods

## 2.1 Chemicals, reagents and enzymes

The source of chemicals, reagents and enzymes for solutions and media which were used in this study, if not specifically stated, were purchased from Merck AG (Darmstadt), Roche Diagnostics (Mannheim), Carl-Roth GmbH (Karlsruhe) and Sigma-Aldrich GmbH (Taufkirchen). The product of restriction enzymes, DNA-modifying enzymes, DNA markers, RNA extraction kit, cDNA synthesis kit, and cloning purpose products were obtained either from New England Biolabs (Schwalbach), JenaBioscience GmbH (Jena), Promega Corporation (Madison, USA), Fermentas GmbH (St. Leon-Rot), or Qiagen (Hilden). Custom DNA oligonucleotides were purchased from MWG Eurofins (Ebersberg) and several antibodies were ordered from Sigma-Aldrich GmbH (Taufkirchen). Fluorescence reagents were obtained from Biotium (Fremont, USA) and Sigma-Aldrich GmbH (Taufkirchen).

## 2.2 Cultivation media

Composition of selective media (Table 1, 2, and 3) are calculated for one liter solved in deionized water. For the solidified media, 1.8 % agar is added to the liquid media. Amino acid supplementation is freshly added after filter-sterilization to the autoclaved medium.

**Table 1. Cultivation media for *S. commune***

| **Cultivation media for *S. commune*** | |
|---|---|
| CYM - Complex Yeast Medium<br>**(Schwalb and Miles, 1967)** | 2 g peptone digested with trypsin<br>2 g yeast extract<br>20 g glucose<br>0.5 g $MgSO_4 \times 7 H_2O$<br>0.5 g $KH_2PO_4$<br>1 g $K_2HPO_4$ |
| CYM-T | CYM with 0.6, 1.2 or 4 mM L-tryptophan |
| MM - Minimal Medium<br>**(Raper and Miles, 1958)** | 5 g glucose<br>2 g aspartic acid<br>1 g $K_2HPO_4$<br>0.5 g $KH_2PO_4$<br>0.5 g $MgSO_4 \times 7 H_2O$<br>120 µg thiamin hydrochloride<br>pH 6.3 (adjusted with NaOH) |
| MM-U | MM supplemented with $10^{-2}$ M Uracil |
| MM-U-T | MM supplemented with $10^{-2}$ M Uracil and 0.6 mM L-tryptophan |
| Top agar | Medium with 2% Bacto-agar |

**Table 2. Cultivation media for *E. coli***

| **Cultivation media for *E. coli* DH5 alpha** | |
|---|---|
| Standard-I Medium, pH 7.5 | 15 g peptone digested with trypsin<br>3 g yeast extract<br>6 g NaCl<br>1 g glucose |

**Table 3. Cultivation media for *S. cerevisiae***

| **Cultivation media for *S. cerevisiae* BY4741** | |
|---|---|
| YPD Medium, pH 7.5 | 10 g yeast extract<br>20 g trypticase peptone<br>20 g glucose |
| YMM | Yeast synthetic drop-out media without Leucine<br>2% Bacto-agar |
| YMM -L | YMM with 1.6 mM Leucine |

## 2.3 Organisms, plasmids, and nucleotides

Organisms and strains that are used in this study are listed in Table 4. The used plasmids (without transient plasmids for sequencing) are listed in Table 5. Oligonucleotides are listed in Table 6, 7, and 8.

**Table 4. Organisms and strains**

| **Name** | **Mating Type and description** | **Source** |
|---|---|---|
| *E. coli* K12 DH5α | F-, *end*A1, *hsd*RA, *sup*E44, *thi*-1, *rec*A1, *gyr*A96, *rel*A1, f80d*lac*ZΔM15 | Gibco Life Technologies (Carlsbad, California) |
| *S. cerevisiae* BY4741 | *MATa; his3Δ 1; leu2Δ 0; met15Δ 0; ura3Δ 0* | Euroscarf (Bad Homburg) |
| *S. commune* 12-43 | Wild type *matA3,5; matB2,3*; *ura-* | Strain Collection |
| *S. commune* H4-8 | Wild type *matA4,6; matB3,2* | Strain Collection |
| *S. commune* 1-69 | Wild type *matA3,22; matB1,2* | Strain Collection |
| *S. commune* 1-106 | Wild type *matA110; matB4,2* | Strain Collection |
| *S. commune* T 50 | *matA76; matB2,2* | Strain Collection |
| *S. commune* 4-39 | Wild type *matA1,1; matB3,2* | Strain Collection |
| *S. commune* T33 | *matA3,1; matB3,2; trp-; ura-* | kindly provided by Kirk Bartholomew, Sacred Heart College, Fairfield/USA |
| *S. commune* EVC1 | *matA3,1; matB3,2;* empty vector control, origin from T33 | This study |
| *S. commune* OEIMP4 | *matA3,1; matB3,2; tef1* promoter fused with *imp* | This study |
| *S. commune* OEIMP6 | *matA3,1; matB3,2; tef1* promoter fused with *imp* | This study |
| *S. commune* 15-2A | *tub* promoter, *Lifeact, eGFP, cdc42* terminator, *phleo*$^{-}$ | Jung *et al.* (2018) |
| *S. commune* F28 | *matA4,6; matB3,2; trp-, Δgap* | Schubert *et al.* (2006) |
| *S. commune Ras1*$^{Q61L}$ (2-1$^{Q61L}$) | *matA4,6; matB1,1; ura-*, *trp-*, *Ras1* constitutive active | Knabe *et al.* (2013) |
| *S. commune Ras1*$^{G12V}$ (T2$^{G12V}$) | *matA4,1; matB2,2; ura-*, *Ras1* constitutive active | Knabe *et al.* (2013) |
| *S. commune Ras1*$^{G12V\_2}$ (2-1$^{G12V}$) | *matA4,6; matB1,1; ura-, trp-, Ras1* constitutive active | Knabe *et al.* (2013) |

**Table 5. Plasmids**

| Name | Description | Origin |
|---|---|---|
| pDrive | PCR cloning vector, poly-A cloning, 3.85 kb, Amp$^{R}$, Kan$^{R}$, | Qiagen |
| pRS415 | 6 kb, binary shuttle vector for *E. coli* and *S. cerevisiae,* Amp$^{R}$, *leu* | Addgene |
| pRSTEFPIMP | 8.5 kb, *tef1* promoter fused with *imp*, pRS415 backbone | This study |
| pTrp | 7.7 kb, *trp,* pBluescript SK+ backbone | Munoz-Rivas *et al.* (1986) |
| pOEIMP | 10.1 kb, *imp* overexpression plasmid, *trp* selection marker, pTrp backbone | This study |

**Table 6. Oligonucleotides-PCR routine**

| Primer | Sequence | Description |
|---|---|---|
| IMP F<br>IMP R | ATGCCCACCGACCTTACCAT<br>CTACTTGGTCGGCTCTGCGT | Inositol monophosphatase |
| TEF1 F<br>TEF1 R | GGTCACCGTGACTTCATCAAG<br>CTTGATGATACCGGTCTCGAC | Translatioon elongation factor 1 |
| M13 F<br>M13 R | GTAAAACGACGGCCA<br>CAGGAAACAGCTATG | Universal primer M13 for cloning purpose |

**Table 7. Oligonucleotides RT-qPCR**

| Primer | Sequence | Description |
|---|---|---|
| ACT F<br>ACT R | GTCCCGCCCTCGAGAAGAGTTA<br>TTGTACTCGTCTCGTGGATA | Actin cytosekeleton |
| TEF F<br>TEF R | AGCTTGGCAAGGGTTCCTTCA<br>AACTTCCAGAGGGCGATATCA | Translatioon elongation factor 1 |
| UBI F<br>UBI R | GAAGGAGTACGATGCGAAGG<br>TCCTCCTCTGCCTTCTTGC | Ubiquitin |
| IMP F<br>IMP R | GAAGCCCGTGCTCGGTG<br>TGGTTGTTGATGCCCTCCGT | Inositol monophosphatase |
| PIS F<br>PIS R | TCGAGCCACAAGACCGTTCA<br>AGGTAGAGGGCGACGAAGAA | Phosphatidyl inositol synthase |
| RAS1 F<br>RAS1 R | CGCACCTCGACGCTTACTCA<br>GTCGAATGTGGAGCGGGATG | Ras1 |
| RAS2 F<br>RAS2 R | ACCAGGATTGGTTCCCCATCG<br>TCTCGATGAAGACGCAGCCA | Ras2 |
| IPPK_2F<br>IPPK 2R | CGGTCGAGATCAAGCCAAA<br>ATGCAGAATCGGCAGGTC | Inositol pentakisphosphate 2-kinase |
| IPMK_2F<br>IPMK 2R | AGCAGTCCCTCGTCCT<br>CTGGCGACGCAGATTCA | Inositol polyphosphate multikinase |

**Table 8. Oligonucleotides for overexpression of *imp***

| Primer | Sequence | Description |
|---|---|---|
| p1forA | **CTAGTTCTAGAGCGGCCGCC**ACCGCCGGAAAAGAACAAGACGTGT* | pRS415 plasmid overhang with *tef1* promoter |
| p1revA | **TAGTCGGCGATGGTAAGGTCGGTGG**GCATTTTGAGTGTTTTCTAAGTGAG* | *tef1* promoter with *imp* overhang |
| p2forA3 | **TCACTTAGAAAACACTCAAA**ATGCCCACCGACCTTACCAT* | *tef1* promoter overhang with imp1 |
| p2revA3 | **CTAAAGGGAACAAAAGCTG**GGACACACGAGGATGACGGTT* | *imp* with pRS415 plasmid overhang |

*overhang is marked in bold

## 2.4 Experiments with *E. coli* DH5α

### 2.4.1 Growth conditions and production of electrocompetent cells

*E. coli* DH5α is electrocompetent cells that is designed for routine cloning applications. *E. coli* DH5α was cultivated in Standard-I medium at 37°C for 16 h (see Table 2). To obtain the electrocompetent cells, a pre-culture was prepared by inoculating *E. coli* DH5α to 20 ml Standard-I medium liquid for overnight. Pre-culture in 50 ml volume was transferred to 500 ml Standard-I medium. The main culture was incubated and shaken at 200 rpm for 3-4 hours at 37°C, until the cells reached an OD 0.7 on Abs 590 nm. The culture was pelleted by centrifugation for 10 min, 4000 rpm, 4 °C. Pelleted cells were washed two times with sterile deionized water and subsequently washed two times with 10 % glycerin. Washing steps were done to wash away salts and materials contained in the medium. *E. coli* cells were dissolved in 2 ml of 10 % glycerin, aliquoted to 100 µl, and stored at -80 °C before usage.

### 2.4.2 Transformation of *E. coli* DH5α

Plasmid DNA was transformed to *E. coli* by electroporation. For each transformation 100 µl electrocompetent cells were thawed on ice. Plasmid DNA was added to the *E. coli* cells. The suspension was filled into an ice-cold electroporation cuvette (electrode distance: 2 mm). Electroporation was done using the Gene Pulser® (Bio-Rad Laboratories Inc., USA) by applying electrical tension of 2.5 kV at 200 Ω and 25 µF. The electroporated suspension was immediately mixed with 900 µl Standard-I medium. The mixture was plated on Standard-I medium supplemented with or without the appropriate selective antibiotic (100 µg/ml Ampicillin or 50 µg/ml Kanamycin). For the PCR cloning vector pDrive, the selection was done in media containing Ampicillin and 40 µg/ml X-Gal (solved in DMSO) as blue-white screening indicator.

## 2.5 Experiments with *S. commune*

### 2.5.1 Cultivation of *S. commune*

*S. commune* was cultivated in Complex Yeast Medium, CYM (Schwalb & Miles, 1967) and Minimal Medium, MM (Raper & Hoffman, 1974) follow by incubation at 25-30°C in the dark or exposure by daylight (see Table 1). Depending on the experiment, *S. commune* was either cultivated in liquid media or solidified media. For the solid culture, a fungal inoculum (0.5 $cm^2$) was excised using a flame sterile metal lancet and directly placed on a solid media. For liquid culture, 4 $cm^2$ agar blocks were excised and transferred into 50 ml medium. The agar block was crushed and homogenized using a sterile laboratory blender (neoLab, Heidelberg) for 30 seconds and then transferred into a 500 ml Erlenmeyer flask. The volume of liquid culture was adjusted to 100-250 ml. Liquid culture was incubated in a laboratory shaker at 30 °C and shaken at 125 rpm.

### 2.5.2 Growth rate measurement and dry weight determination

The growth rate of *S. commune* was calculated from the mycelia extension rates. Measurement of the mycelium was performed in the same area which was marked using a permanent marker. Measurement was performed every day until the fungus reached maximum growth on plate. Growth rate analysis was done by calculating the average of three times measurements of the mycelial diameter. To determine the dry weight, a fungus agar block was inoculated over the sterile cellophane on the agar plate. After several days of inoculation, grown mycelia were peeled from the cellophane layer and placed on the weighing dishes. Mycelia were dried using a drying oven (WTC Binder, Binder GmbH, Tuttlingen) at 80°C for 6 hours and weighed using an analytical balance (Satorius Basic, Satorius AG, Göttingen). Three replicates of each treatments were applied to this experiment. Significant difference between the control and the treatment were determined by two side T-test ($p$ value $\leq 0.05$).

### 2.5.3 Rapid DNA isolation

A quick and non-laborious fungal DNA isolation (Cenis, 1992) was performed using either a plate culture or liquid culture. 1 mg of mycelial powder from liquid culture or 10 $mm^2$ mycelial mat from a plate culture was transferred into a 1.5 ml reaction tube containing 300 µl of DNA extraction buffer (200 mM Tris-HCl, pH 8.0; 25 mM EDTA; 250 mM NaCl; 0.5 % SDS). In case of mycelial mat sample, mycelia were crushed using a metal lancet or plastic pestle. Fungal extract and buffer mix were homogenized with a vortexer. 150 µl of 3 M sodium acetate, pH 5.2 were added to the mix follow by incubation for 10 min at -20 °C. After centrifugation at 13000 rpm for 5 min, the supernatant was mixed with 0.5 vol of ice-cold 2-propanol in a new reaction tube. Precipitation of DNA was done by centrifugation of a tube at 13000 rpm for 25 min. Supernatant was aspirated and pellet washed with 70 % ethanol and then dried for ca. 2

min in SpeedVac (Savant™ DNA SpeedVac™, Thermo Fisher Scientific, Schwerte). The DNA pellet was solved with 50 µl sterile deionized water and stored at -20 °C.

### 2.5.4 DNA isolation using CTAB

DNA extraction (Murray and Thompson, 1980) using a surfactant CTAB (Cetyl trimethyl ammonium bromide) was selected, to yield a high concentration of total cellular DNA (1-2 µg/µl). Fungal liquid culture was prepared to target a high concentration DNA. Removing exopolysaccharide from liquid culture can be tedious, therefore CTAB was used to remove the fungal exopolysaccharides. CTAB binds to exopolysaccharides in the presence of high salt concentration, removes membrane lipids and promotes cell lysis, thus resulting in liberation of total cellular nucleic acid. Mycelia were harvested from 300 ml liquid culture. After 2 days cultivation, liquid media were removed with a nylon net and the mycelia was rinsed with deionized water. This procedure was repeated at least 3 times until the mycelia were completely washed away from the remaining liquid media and exopolysaccharide. Wet mycelia were ground immediately with liquid nitrogen using mortar and pestle. A 50 ml reaction tube was immersed in liquid nitrogen and 1 g frozen fungal powder was transferred immediately into the tube. It is important to notice that the mycelia had to remain frozen before DNA extraction buffer (0.7 M NaCl; 100 mM Tris/ HCl pH 8.0; 10 mM EDTA) was added. DNA extraction buffer was pre-warmed at 65°C for 15 min. Volume of 10 ml DNA extraction buffer was added to the tube, followed by 60 min incubation at 65°C and 1 ml of CTAB-NaCl solution (10 % CTAB; 0.7 M NaCl) was added subsequently. The tube was shaken gently, followed by incubation at 65 °C for 15-30 min. The mixture was chilled on ice and one volume of chloroform-isoamyl alcohol was added to the supernatant. The tube was gently shaken, incubated on ice bath for 5 min, and centrifuged at 7000 rpm for 10 min, 4°C. The upper aqueous phase (clear supernatant) was taken out using 5 ml pipette and transferred into a new 50 ml reaction tube. This procedure was done carefully to ensure that only the clear supernatant was aspirated, without disturbing the interphase. Extraction of interphase using DNA extraction buffer, CTAB-NaCl solution, and chloroform-isoamylalcohol was repeated three times. DNA was precipitated by adding 0.7 volume of isopropanol and followed by overnight incubation at 4°C. Precipitated DNA was centrifuged and the supernatant was discarded. DNA pellet was washed with 70 % ethanol and dried with a vacuum (Savant™ DNA SpeedVac™, Thermo Fisher Scientific, Schwerte). A transparent DNA pellet was eluted in 100-500 µl TE buffer (10 mM Tris-HCl, pH 8.0; 1 mM EDTA) or sterile deionized water. DNA was transferred to a 0.5 ml reaction tube and stored at -20°C for up to 6 months.

### 2.5.5 RNA isolation

Fungal cultivation was performed on solid medium-cellophane membrane. Amount of 100 mg powdered mycelia was harvested by grinding with liquid nitrogen. The procedure was performed based on the manufacturers guideline RNeasy Plant Mini Kit (Qiagen, Austin, USA).

Buffer RLC mixed with β-mercaptoethanol was prepared befpre the RNA isolation was carried out. Before RNA elution from the column, 3 min incubation at RT was carried out. RNA was eluted from the spin column using 50 µl provided RNAse-, DNAse- free water.

### 2.5.6 Quality, purity, and quantitation for DNA and RNA

DNA/RNA visualization using 1% agarose gel was performed to determine DNA/RNA quality. For this purpose, agarose powder was suspended in 1 x Tris Acetate-EDTA (TAE) buffer (diluted from 10 x TAE, Carl Roth, Karlsruhe) and boiled in the microwave. Gel was casted using an electrophoresis apparatus and left until vitrified. The solidified gel was placed and completely immersed on the electrophoresis chamber containing 1 x TAE running buffer. It is important to notice, that the buffer should cover the gel surface. DNA or RNA in volume of 3-5 µl was mixed with 2 µl loading dye and loaded onto the well of solidified gel. On the separate well, 2-log DNA marker (New England Biolabs, Schwalbach) in volume of 5 µl was loaded into the gel. Gel electrophoresis was performed at 100 V, 0.02 mA for 1 h using an electrophoresis apparatus (Scie plas PSU 400/600, Carl Roth, Karlsruhe). After electrophoresis, agarose gel was stained in 1 µg/ml ethidium bromide solution for 15 min. Gel was visualized using Infinity gel documentation imaging system and software Infinity-Capt Version 14.1a.

The purity and concentration of DNA/RNA was analysed using DeNovix DS-11 Spectrophotometer (Biozym, Hessisch Oldendorf). The purity and concentration were determined automatically based on the Lambert Beer Law.

## 2.6 Inositol monophosphatase activity measurement

### 2.6.1 Crude extraction of *S. commune*

Five to seven days old liquid culture of *S. commune* was ground with liquid nitrogen. Volume of 4 ml crude extraction buffer (25 mM Tris/HCl; 1 mM DTT; 1 mM EDTA, pH 7.4) was added to 1 g of mycelia powder. Incubation on ice was performed for 15 min. The reaction mix was centrifuged at 14000 rpm at 4 °C for 10 min (Mikro200R, Hettich Zentrifugen, Tuttlingen) and the supernatant containing total crude was either continued to downstream procedure, or stored at -80 °C.

### 2.6.2 Protein quantitation

Determination of crude/protein concentration was performed according to Bradford assay (Bradford, 1976). Bovine Serum Albumin (BSA) as protein standard (Sigma-Aldrich GmbH, Taufkirchen) was prepared within the concentration range 0 to 1.5 mg/ml. Crude/protein was diluted 10-100 times, if necessary.

The assay was performed using a 96 well-plate. The assay cocktail comprise of 5 µl of standard or crude/protein and 250 µl of Bradford reagent (Sigma-Aldrich GmbH,Taufkirchen). The assay cocktails were incubated for 5 min at RT. The maximum absorption of the assay cocktail was determined at 595 nm wavelength (UltrospectTM 2100 Pro UV/Visible Spectrophotometer, Amersham Biosciences). Crude/protein concentration was determined by comparing the net of Abs 595 value against the standard curve.

### 2.6.3 Malachite Green assay

IMPase activity was determined as lithium-sensitive phosphate release measured using the malachite green reagent. The assay was adapted from Kalujnaia *et al.* (2010). Inorganic Phosphate (Pi) standard was obtained from serial dilutions of a 1 M $Na_2HPO_4$ stock (0-100 nmol/ml). Crude extract was diluted to 1 µg/ml in Buffer A (25 mM HEPES; 0.25 M sucrose; 2 mM MgCl2; 1 g/ml protease inhibitor cocktail, pH 7.4 (Sigma-Aldrich GmbH) and Buffer A containing 50 mM LiCl. The reaction was mixed and incubated on ice for 10 min. Volume of 10 µl of Pi standard or enzyme solution (with and without LiCl) was added to the 96 well-plate. Volume of 90 µl Buffer B (25 mM HEPES; 2 mM $MgCl_2$; 200 µM inositol monophosphate, pH 7.4) was added to the Pi standards and enzyme solutions. The assay cocktail was incubated either 15, 30, 45, 60 or 120 minutes at RT. The reaction was terminated by applying 50 µl of freshly prepared Malachite Green reagent (10 ml Malachite Green acidic solution; 2.5 ml of 7.5% Ammonium molybdate; 0.2 ml of 11% Tween 20) (Baykov *et al.*, 1988) to the assay cocktail. Malachite Green acidic solution (60 ml of 1.84 g/L $H_2SO_4$, 300 ml deionized water, 0.44 g malachite green) can be prepared and storage for 1 year. The maximum absorption to determine phosphate release was performed at 620-640 nm wavelength (UltrospectTM 2100 Pro UV/Visible Spectrophotometer, Amersham Biosciences).

## 2.7 Protoplast preparation and transformation of *S. commune* an

The protoplast preparation and transformation of *S. commune* was done after some modification from one day old 300 ml liquid cultures were prepared by crushing and homogenizing two days old 100 ml fungal liquid pre-culture. Liquid culture was divided and transferred to several 50 ml reaction tubes. Mycelia were harvested by centrifugation (Megafuge 1.0R, Hettich centrifuge) at 2500 rcf for 10 min. Medium was discarded and pelleted mycelia were washed with 10 ml of 0.5 M $MgSO_4$, pH 6.3 adjusted with 1 M MES). Washing step was repeated two times. Wet mycelia were mixed with 5-10 ml lysing enzyme solution (100 mg lysing enzyme of *Trichoderma harzianum* and 100 mg ß-glucanase from *Trichoderma longibrachiatum*, Sigma-Aldrich GmbH, Taufkirchen in 100 ml $MgSO_4$ pH 6.3). The lysing enzyme-mycelia mixture was gently shaken at 60 rpm, 30°C for 3-5 h. While incubated, protoplasts formation was monitored by sampling a few microliters of the mixture and checking under microscopy after 3 h of incubation. The enzyme-mycelia mixture was filtered using a sterile Miracloth filter tissue (Merck Millipore) to separate the protoplasts from the mycelia. One volume of 1 M sorbitol was added to the protoplasts and incubation was allowed for 10 min at RT. The mixture was centrifuged at 2500 rcf for 10 min. Supernatant was removed carefully without interfering the bottom layer containing protoplasts. Volume of 5 ml protoplasts was left in each reaction tube. From all tubes, each 5 ml protoplast mix were combined and collected in a single reaction tube. Supernatant was removed very carefully by pipetting. Volume of 5 ml of 1 M sorbitol was added to the tubes. The tubes were centrifuged at 2500 rcf for 10 min at RT. Washing step with sorbitol was repeated one time. After all, protoplasts were solved in 1 M sorbitol supplemented with 50 mM $CaCl_2$. Protoplasts were counted using a thoma chamber and stored on -20°C for one week if it was not proceeded to transformation.

Transformation was performed with 100-200 µl protoplasts and 1-20 µg plasmid DNA. The ideal ratio for transformation is 1: 10 (plasmid DNA: protoplasts). Plasmid DNA and protoplast were mixed gently using a pipette tips and incubated on ice bath for 15 to 30 min. One volume of freshly made PEG solution (2 g PEG4000 in 2 ml 10 mM Tris HCl, 10 seconds in microwave to solve and sterilize) was added and mixed gently followed by incubation for 5 min at RT. Protoplasts were incubated in regeneration media (CYM liquid supplemented with 0,5 M sucrose, with or without respective selection marker) for 24-48 h, at RT. Regenerated protoplasts were embedded into a lukewarm top-agar with selective antibiotics (e.g. phleomycin, nourseothricin) or marker (e.g. L-tryptophan, uracil). Growth of selective mycelia was observed for 3-14 days and growing mycelia was selected on a second and third media containing respective antibiotics or without selection marker. Putative transformants were further screened and verified by PCR using specific primer pairs.

## 2.8 Construction plasmid: *imp* overexpression for *S. commune*

A constitutive and strong promoter, *tef1* promoter (Gola and Kothe, 2003) was employed to construct overexpression system of *imp*. The gene of interest *imp* (1497 bp) was fused with the *tef1* promoter (985 bp) by yeast recombination method (Gietz *et al.*, 1995; Gietz and Sugino, 1988) using a binary shuttle plasmid pRS415 (Figure 7), resulting in pRSTEFPIMP (Figure 9). The fusion of *tef1* promoter and *imp* later was cloned to the pTrp containing tryptophan marker for selection, followed by transformation in *S. commune*.

### 2.8.1 Yeast Recombination

In this study, yeast recombination protocol was adapted from and slightly modified after Freihorst (2015). *S. cerevisiae* BY4741, Δ*LEU* was opted for the yeast recombination, thus the leucin was used as a marker to select positive transformants. A set of primers was designed specifically for yeast recombination purpose (Table 8). The sequences of *tef1* promoter and the *imp* gene were amplified using two specific primer pairs that possesses overhangs 20-25 base pairs to the fragments which need to be fused (Figure 8). Primer sequences and recombination scheme are listed in Table 8. The two fragments, *imp* and *tef1* promoter were amplified using a proofreading, high-fidelity DNA Polymerase Q5 (New England Biolabs, Schwalbach) to avoid incorrectly-paired nucleotides.

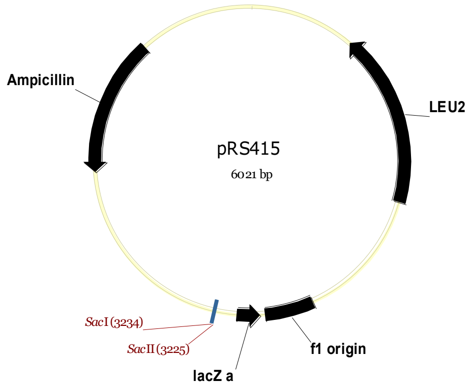


Figure 7. A binary shuttle plasmid pRS415 (A) used for yeast recombination to clone a fusion fragment *tef1p* and *imp* gene. The plasmid pRS415 encompasses multiple sites for cloning located before *lacZ a* operon. It possesses LEU2 (leucin), a marker gene for yeast cloning and a marker gene *bla* (Ampicilin) for *E. coli* cloning. Two specific restriction sites (SacI and SacII) used for linearization to clone the fusion fragments *tef1p* and *imp* gene.

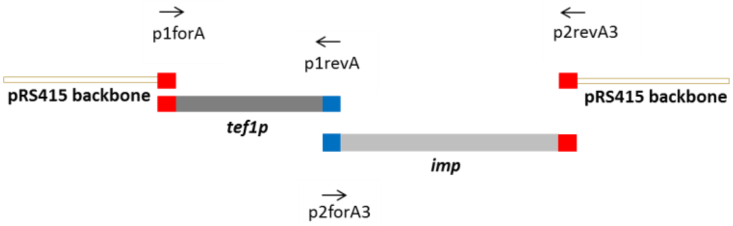


Figure 8. Recombination in *S. cerevisiae* was carried out using pRS415 plasmid. Fusion product of *tef1* promoter (*tef1p*) and *imp* gene. The overlapping sequences (red box) were created using primers, that possesses overhangs from pRS415 backbone (SacII restriction site for p1forA for and SacI restriction site for p2revA3). Overlapping sequences between two adjacent fragments *tef1p* and *imp* (blue box) was created using overhang primers p1revA and p2forA3.

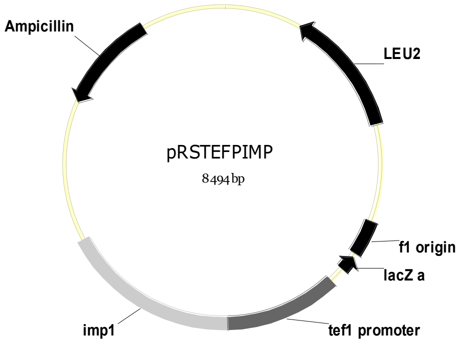


Figure 9. Fusion fragments *tef1p* and *imp* were successfully clone to plasmid pRS415, which later given a name as pRSTEFPIMP.

### 2.8.2 Cultivation of *S. cerevisiae* BY4741, Δ*LEU*

*S. cerevisiae* BY4741, Δ*LEU* was pre-cultivated in 5 ml liquid YPD medium overnight. The culture was shaken in the environmental incubator at 150 rpm, 30°C. The next day, 2 ml of pre-culture was added to 50 ml liquid YPD medium, followed by incubation in a laboratory shaker at 30° C. The optical density was measured at 600 nm (OD 600) and the growth of yeast was monitored every hour after 2 hours. Yeast was harvested by centrifugation at 4000 rpm, 10 min, when it has reached OD600 = 0.3, which was between 3 – 4 hours. Transformation of yeast was carried out using a fresh yeast cells before it has reached its stationary growth. In this phase, the cells have the highest competence to take up plasmid DNA. After harvesting, yeast cells were washed three times using pre-chill, sterile water and resuspended in 1 ml lithium acetate solution (100 mM lithium acetate in 10 mM Tris/HCL pH 8.0 and 1 mM EDTA). The resuspended cell was transferred to 1,5 ml reaction tube and the yeast transformation was performed. The storage of competence yeast cell is possible at -20°C only up to 1 month.

### 2.8.3 Yeast transformation

Yeast recombination was carried out using 100 µl of yeast cells, 100 ng plasmid pRS415, 50 ng purified PCR product *tef1p*, 50 ng purified PCR product *imp,* 10 µg ss (single stranded) salmon sperm DNA and 600 µl PEG solution (50 % PEG 4000 in 100 mM lithium acetate). The plasmid pRS415 were isolated from *E. coli* DH5α using a kit GeneJET Plasmid Miniprep (Thermo Fisher Scientific). Fragments *tef1p* and *imp* were amplified using a high-fidelity DNA polymerase Q5 and the PCR product was visualized in 1% agarose electrophoresis. The right product for each fragment was excised and promptly purified using GeneJET Gel Extraction Kit (Thermo Fisher Scientific). Sheared ss salmon sperm in a relative high concentration was employed to overcome the cell wall binding with plasmid DNA, allowing a high uptake of exogenous plasmid DNA by the yeast cell.

The component for yeast recombination was mixed gently with a pipette for 15 seconds and promptly incubated for 30 min at 30 °C. After incubation, 70 µl DMSO were added to the mix and tube was inverted several times and 15 min incubation at 42 °C and tube was chilled 1 min on ice. Yeast cells were spun down at 13000 rpm for 15 seconds. The yeast pellet was solved in Yeast Minimal Medium (in the absence of leucine, YMM -leu) and 250 µl were plated directly on YMM-leu solid media. Positive and negative control were prepared to avoid false positive outputs. 100 ng circular plasmid pRS415 was added to the mix for the positive control, and 100 ng linearized plasmid pRS415 (double digestion using endonucleases SacI and SacII) was added to the mix for the negative control, instead of 100 ng linearized plasmid pRS415. Selection was done on YMM -leu solid media. Yeast colonies were grown after 2-5 days and putative transformants were further screened and validated.

### 2.8.4 Screening and validation of recombined product

To screen the putative transformants, total yeast DNA (including genomic and plasmid DNA) was isolated using a protocol from Colot and co-worker (Colot *et al.*, 2006). Total DNA was isolated directly from a YMM-leu plate containing transformed yeast cells. Volume of 1 ml resuspension solution (50 mM glucose, 25 mM Tris/HCl, 1 mM EDTA, pH 8.0) was added to the agar and yeast colonies were scraped softly with a flame-sterile triangle head metal spreader. The resolved yeast solution was transferred to a 1.5 ml reaction tube using a pipette and subsequently centrifuged at 13000 rpm for 15 seconds. Supernatant was dispensed and 200 µl lysis buffer (2 % Triton X-100, 1 % SDS, 100 mM NaCl, 10 mM Tris/HCl, 1 mM EDTA), 0.3

g sterilized acid wash glass beads, and 200 µl phenol/chloroform/isoamyl alcohol were added to the pellet. The cell suspension was disrupted vigorously using a vortexer for 2-3 min and pelleted by centrifugation at 13000 rpm for 5 min. A volume of 100 µl supernatant containing total DNA was transferred into a new reaction tube and one volume of 0.6 M sodium acetate, pH 5.2 was added to the supernatant. The reaction tube was inverted 3-5 times. 2.5 Vol of 96 % ethanol was added to the mix and the tube was straightaway centrifuged at 13000 rpm for 5 min. Obtained pellet was washed with 70 % ethanol and air dried. The whitish-transparent DNA pellet was solved in 50 µl deionized water. 1 µl of total DNA containing putative plasmid DNA encompassing fusion of *tef1p* and *imp* gene was cloned to *E. coli* DH5α (see this section). The positive colonies were selected on Standard-I containing ampicillin were screened with PCR. Every single putative positive colony was selected (marked with number) and grown on Standard-I containing ampicillin. PCR was done using M13 standard primer pairs and DreamTaq polymerase (Thermo Fisher Scientific) to check existence of the recombined fusion product. Acquired positive colonies which were revealed by PCR, were afterwards cultivated on Standard-I and miniprepped. Plasmids containing *tef1* promoter and *imp* gene fusion was named pRSTEFPIMP (8,5 kb). Restriction pattern of pRSTEFPIMP was examined using restriction endonucleases PciI and HindIII before sending to sequencing.

### 2.8.5 Cloning of *tef1* promoter and *imp* gene to pTrp

After *tef1* promoter and *imp1* gene were successfully integrated to pRS415, the fusion product containing *tef1* promoter and *imp1* was excised with restriction enzyme HindIII and PciI. The resulted fragment (2.9 kb) was cloned to the HindIII and PciI site in pTrp with T4 ligation (New England Biolabs). The ligation mix was prepared according to the manufacturer's protocol with modification. The plasmid pTrp was linearized using using endonucleases HindIII and PciI, resulting sticky-end sequences. The ligation mix contained 50 ng linearized plasmid pTrp, 40 ng DNA fragment (*tef1p* and *imp* gene), 10 µl 2x T4 DNA ligase reaction buffer, 1 µl quick ligase, and nuclease-free water which was adjusted to the end volume 20 µl. The reaction was gently mixed by pipetting and spun down briefly. The ligation mix was incubated at 16°C, overnight. On the next day, 5 µl of the ligation mix was proceeded to transformation of *E. coli* (see section 3.1.2). The resulted plasmid was called pOEIMP.

## 2.9 Proteome analysis using 2D-PAGE

### 2.9.1 Cytosolic protein isolation

Mycelium was harvested and ground in liquid nitrogen. Cytosolic protein isolation was modified after Bordier (Bordier, 1981). Amount of 5 g powdered mycelium was transferred into a 50 ml reaction tube, filled up to 25 ml with solution A and mixed. Afterwards 10 ml solution B was added and mixed by inverting the tube. The sample was incubated on ice for one hour. In between incubation time, tube was shaken every 20 min. The tube was centrifuged at 11000 rpm, 4 °C for 15 minutes. Supernatant containing cytosolic protein was decanted into a new reaction tube and incubated at 30 °C for three minutes follow by centrifugation at 1600 rpm, RT for 10 min. The aqueous phase was transferred into a new reaction tube, filled up with one volume of precipitation buffer and incubated at 4 °C overnight. After incubation, the precipitated protein was centrifuged at 11000 rpm, 4 °C for 10 min. Supernatant was discarded, and pellet was washed with acetone for four times. Last washing step with the acetone was done in a 2 ml reaction tube. A hardened pellet from the fourth washing was completely transferred to a 2 ml reaction tube and mixed with the acetone. The pellet was dried out in a vacuum centrifuge and resuspended in 0.5 ml rehydration buffer. Sample was centrifuged at 13,000 rpm, 4 °C for 10 min. Supernatant was transferred into a new reaction tube. Protein concentration was quantified using Bradford assay (Bradford, 1976).

### 2.9.2 First and second dimension of 2D gel electrophoresis

In the first dimension, protein was separated based on their isoelectric point. The amount of 250 µg protein was applied onto an Isoelectric Point Gradient-strip, IPG-strip (Immobiline DryStrip pH 3 - 7 NL; pH 3 - 11 NL, 24 cm, GE Healthcare, Uppsala, Sweden). 2 % (v/v) of IPG buffer (pH 3-11 NL) and 5.4 µl of DeStreak reagent (GE Healthcare, Uppsala, Sweden) were added to the rehydrated protein. The volume of protein samples was adjusted to 450 µl using a rehydration buffer.

**Table 9. Buffer and solutions for 2D-PAGE**

| **Buffer and solutions for 2D-PAGE** | |
|---|---|
| Running gel buffer<br>pH 8,8 adjusted with 10M HCl | 182 g Tris/HCl<br>4 g SDS |
| Polyacrylamide gel (for 6 gels) | 247.5 ml polyacrylamid (37.5: 1)<br>165 ml running gel buffer<br>33 g glycerine<br>33 µl TEMED<br>3 ml 10 % APS |
| 10x Running buffer | 25 mM Tris<br>192 mM glycin<br>0.1 % SDS |
| Blue agarose | 1 % agarose in running gel buffer<br>a spatula tip Coomassie Brilliant Blue |

Protein mixtures were transferred to a reswelling tray (GE, Healthcare, and Uppsala, Sweden) and IPG-strip were laid on top of the protein mixture. IPG-strip was left overnight at RT. A day after, the stripes were laid on top of a ceramic tray (Ettan™ IGPhor II™ focusing device, GE Healthcare, Uppsala, Sweden) with the gel facing on top. The stripes were covered with Immobiline DryStrip cover fluid (GE Healthcare, Uppsala, Sweden). The isoelectric focusing was programmed as following: 300 V for 4 h (gradient), 600 V for 4 h (gradient), 1000 V for 4 h (gradient), 8000 V for (gradient) and 8000 V for 24000 Vhr (step). For 2D gel electrophoresis, the strips were incubated for 15 min in equilibration buffer 1 (6 M urea, 2 % SDS, 30 % glycerine, 3.3 % separating buffer, 1 % DTT) and 15 min in equilibration buffer 2 (6 M urea, 2 % SDS, 30 % glycerine, 3.3 % separating buffer, 24 % iodacetamide). Strips were loaded on top of 10 % (w/v) polyacrylamide gels. Buffers and solutions which are required to prepare polyacrylamide gels are listed in Table 9. Gel electrophoresis was performed using Ettan™ DALTtwelve electrophoresis unit (GE Healthcare, Uppsala, Sweden). The lower chamber was filled with 1x SDS running buffer until the limit line (marked with a sticker in the outer part of chamber) and 2x SDS running buffer was added to the upper chamber. Gels were run at 1 W per gel for 1 hour and at 15 W per gel for 8-10 hours.

After 2D gel electrophoresis, each gel was soaked in the fixation solution (40% methanol, 7% acetic acid) for 30 minutes and stained with staining solution (20 % methanol, 2 % o-phosphoric acid, 10 % ammonium sulfate, 0.1 % Coomassie Brilliant Blue G-250) overnight. Gels were neutralized with Tris-base pH 6.5 (adjusted with 85 % O-phosphoric acid) for 10 min and destained with destaining solution (25 % methanol). Gels were scanned with an Epson Bio Step ViewPix scanner and subsequently analyzed with the Delta2D imaging software v. 4.3 (Decodon, Greifswald, Germany).

### 2.9.3 Protein/ peptide analysis using MALDI TOF/MS-MS

Proteins/peptides were analyzed using matrix-assisted laser desorption ionization times-of-flight mass spectrometry (MALDI TOF/MS). Tryptic digestion of protein spots was done as according to Shevchenko *et al.* (1996) with several alterations. Protein spots were excised and incubated in 200 µl of 50 mM $NH_4HCO_3$ : acetonitril (1:1) solution for 30 minutes and then in 100 µl acetonitrile shortly. In this step, the gels were shrunken in size. Following 5 minutes incubation in 100 µl of 50 mM $NH_4HCO_3$ and 15 min incubation in 100 µl acetonitrile were performed. Before next solution was added, the previous solution was always discarded using a pipette. It is important to notice that the gel would be rehydrated after $NH_4HCO_3$ was added. After the last solution was removed, protein spots were vacuumed-dried. The protein spots were digested with 10 µl trypsin solution (20 µg/µl trypsin (Promega, Mannheim, Germany) for 40 min on ice. After removal of the enzyme solution, a volume of 3 µl of 25 mM $NH_4HCO_3$ was added to the protein spots and incubated at 37 °C overnight. Peptides were extracted with 10 µl extraction buffer (25 mM $NH_4HCO_3$ and 0.1 % TFA/acetonitrile, 1:1). To identify the peptide, 1 µl HCCA matrix (10 mg/ml alpha cyano-4-hydroxy-cinnamic acid in 30 % acetonitrile, 70 % distilled water with 0.1 % trifluoroacetic acid) was transferred to an Anchor Chip Target (Bruker Daltonics, Bremen) and mixed with 1 µl of the tryptically digested sample. After sample crystallization, proteins were analyzed by an ultrafleXtreme™ MALDI-TOF/TOF device (Bruker Daltonics, Germany). Sample measurement was set up with flexControl software followed by analysis with flexAnalysis (both Bruker Compass 1.3). Protein peaks were analyzed, and database search was done using the published genome sequence *S.*

*commune* v3.0. Search parameters comprised of oxidized methionine as variable modification, carbamidomethyl cysteine as fixed modification, one mis cleavage and a peptide mass tolerance of 100 ppm and a fragment mass tolerance of 0.6 Da. Significant results were determined according to the Mascot score with an allowed likelihood for a random hit of $P$-value ≤ 0.05.

## 2.10 Signal protein analysis

### 2.10.1 Signal protein isolation and purification

Cellular protein and membrane associated signaling proteins were isolated from mycelia with ReadyPrep protein extraction kit – Signal (Bio-Rad Laboratories Inc., USA).

**Table 10. Buffer and solutions for PAGE and LC-MS**

| **Buffer and solutions for PAGE and LC-MS** | |
|---|---|
| 10% Separating gel | 4.1 mL deionized water<br>3.3 ml Acrylamide (30% 37.5:1)<br>2.5 ml 1,5 M Tris/HCl, pH 8.8<br>100 µL 10% SDS<br>10 µL *N*,*N*,*N*′,*N*′-tetramethylethylene-diamine (TEMED)<br>32 µL 10% Ammonium persulfate (APS) |
| 4% Stacking gel | 6.1 mL deionized water<br>1.3 ml Acrylamide (30% 37.5:1)<br>2.5 ml 1,5 M Tris/HCl, pH 6.8<br>100 µL 10% SDS<br>10 µL *N*,*N*,*N*′,*N*′-tetramethylethylene-diamine (TEMED)<br>100 µL 10% Ammonium persulfate (APS) |

Protocol was done according to the manufacturer's manual which was adapted for *S. commune*. Volume of 1 ml buffer S1 was added to 0.5 g frozen powder mycelia. The suspension was sonicated on ice for 40 sec bursts, pulse 01; 01, amplitude 20%, repeated 5 times. In between the sonication, the lysate was chilled on ice for 1 min (ultRasonic processor VCX 130 PB (Sonics & Material Inc., Newtown, CT, USA). After sonication, volume of 0.5 ml chilled buffer S2 was added into the lysate. The suspension was vortexed using a multitube vortexer mixer for 1 min and the vortexing procedure was repeated 5 times. In between vortexing procedure, the lysate was chilled on ice for 1 min. The tube was centrifuged at 14.000 rpm, 4°C for 20 min (Mikro 200 Hettich Zentrifugen, Tuttlingen). Supernatant was discarded using a pipette. The tube was placed on ice bath. Pellet was suspended with 170 µl chilled buffer S1. The mixture was vortexed until the pellet was completely resolved. Volume of 170 µl chilled buffer S2 was added to the suspension. The suspension was vortexed using a multitube vortexer mixer for 1 min and the vortexing procedure was repeated 5 times. In between vortexing procedure, the lysate was chilled on ice for 1 min. The tube was centrifuged at 14.000 rpm, 4°C for 20 min. The final pellet containing signal protein was resuspended using 0.2-0.3 ml PSB. The tube was vortexed 5 times and rested for 1 min on ice bath in between vortexing step. The sample was centrifuged at 14000 rpm for 10 min at RT. Protein concentration was determined using Bradford assay (see procedure in the section 2.6.2).

Amount of 50 µg protein were loaded into 10% SDS PAGE and electrophoresis was run without separation (see Table 10). This step was necessary to perform to remove the PSB buffer. After electrophoresis, gels were washed two times with ultrapure Milli-Q® Water. Protein bands were cut into little cubes (approximately 1 mm$^3$) and transferred to a 1,5 ml tube. In-gel digestion was performed after Shevchenko et al. (1996) using 50 ng/µl trypsin-LysC, Mass Spec Grade (Promega, Mannheim, Germany) in 50 mM $NH_4HCO_3$. Peptides were extracted with trifluoroacetic acid (TFA) and acetonitrile (in different concentration 50 %, 70 %, and 90 %, respectively) and were purified with 10 kDa MWCO filter (VWR European, Langenfield, Germany). Dried peptides were solubilized in 0.05% TFA in 2:98 acetonitrile/water (v/v) and subjected to LC-MS/MS analysis.

### 2.10.2 LC-MS/MS analysis and protein database search

This protocol was kindly provided by Thomas Krüger (Hans Knöll Institute, HKI, Germany). Protein/peptide analysis was performed using LC-MS/MS analysis on an Ultimate 3000 nano RSLC system connected to a QExactive HF mass spectrometer (both Thermo Fisher Scientific, Waltham, MA, USA). Initial peptide trapping for 5 min on an Acclaim Pep Map 100 column (2 cm x 75 µm, 3 µm) at 5 µL/min was followed by separation on an analytical Acclaim Pep Map RSLC nano column (50 cm x 75 µm, 2µm). Mobile phase gradient elution of eluent A (0.1% (v/v) formic acid in water) mixed with eluent B (0.1% (v/v) formic acid in 90/10 acetonitrile/water) was performed as following steps: 0 min at 4% B, 5 min at 5% B, 15 min at 6% B, 100 min at 8% B, 150 min at 12% B, 250 min at 23% B, 300 min at 34% B 320 min at 41% B, 340 min at 52% B, 350 min at 60% B, 360 min at 75% B, 365-375 min at 96% B, 375.1-400 min at 4% B. Positively charged ions were generated at spray voltage of 2.2 kV using a stainless steel emitter attached to the Nanospray Flex Ion Source (Thermo Fisher Scientific). The quadrupole/orbitrap instrument was operated in Full MS / data-dependent MS2 (Top10) mode. Precursor ions were monitored at m/z 300-1500 at a resolution of 120k FWHM using a maximum injection time (ITmax) of 100 ms and an AGC (automatic gain control) target of 1e6. HCD fragmentation at 30% normalized collision energy (NCE) generated MS2 ions, which were scanned at 15k FWHM (ITmax=100 ms, AGC= 2e5). Dynamic exclusion of precursor ions was set to 35 s. The LC-MS/MS instrument was controlled by Chromeleon 7.2, QExactive HF Tune 2.8 and Xcalibur 4.0 software.

The obtained tandem mass spectra were searched against the JGI database of *S. commune* v.3 (2017/02/02) using Proteome Discoverer (PD) 2.1 (Thermo) and the algorithms of Mascot 2.4 (Matrix Science, UK), Sequest HT and MS Amanda. Two missed cleavages were allowed for trypsin digestion. The precursor mass tolerance was set to 10 ppm and the fragment mass tolerance was set to 0.02 Da. Modifications were defined as dynamic Met oxidation and static Cys carbamidomethylation. At least two peptides per protein and a strict false discovery rate

(FDR) < 1% (reverse decoy) were required for positive protein hits. Label-free quantification of the Top 3 unique peptides per protein was based on the precursor ion area detector approach implemented in PD 2.1. Quantitation data was normalized using the total peptide amount approach.

## 2.11 Microscopy

### 2.11.1 Stereomicroscopy and fluorescence microscopy

Fruiting body was examined directly using a stereomicroscope to allow three-dimensional visualization. On the other hand, fluorescence microscope was used to examine hyphae, mycelia, and spores. Samples were prepared using a cover slip culture. A sterile cover slip was placed on the surface of the solid medium. Several fungal agar blocks in size 5 $mm^2$ were placed in four sides or corners of a cover slip. Mycelia could be stained after 3 - 6-day post inoculation (dpi). To visualize fungal cell wall, mycelia were stained with Calcofluor-white (CW), a fluorescent dye that binds strongly to structures containing cellulose and chitin. A drop of 10 µg/ml CW (in PBS) was placed on an object glass slide. After mycelia were grown on a cover slip, the cover slip was placed on top of the object slide that has a drop of CW. Stained mycelia were examined with a fluorescence microscope using a blue filter (02), which is excited and emitted at 358 and 463 nm (Axioplan2, Carl Zeiss MicroImaging, Jena, Germany). The similar procedure was repeated for nuclear staining using 50 µg/ml 4'-6-Diamidino-2-phenylindole (DAPI, solved in PBS) and examined with a fluorescence microscope using a filter 02 (excited and emitted at 358 and 463 nm, correspondingly). Images from both microscopies were taken with a digital camera Insight Firewire 4 image sample (Diagnostic Instruments, Sterling Heights, MI) and processed with SPOT imaging software (Diagnostic Instruments, Sterling Heights, MI). To examine the spores, a fungal culture plate which fruiting body has grown, was placed upside down, thus spores could be collected on a lid of petri dish. Spores were stained using fluorescence dye CW or DAPI in a similar manner to mycelia.

### 2.11.2 Confocal Laser Scanning Microscopy

Time lapse was necessary performed to observe the actin dynamic and vesicles dynamic. The Lifeact GFP strain was cultivated in a 24 well plate using a half concentrated CYM solid. Confocal microscopy was performed with Zeiss LSM 780 (Jena, Germany) and 40x/1,30 NA EC Plan-Neofluar objective. For eGFP and FM4-64 visualization, Ar/ML 488/514 nm Laser with 0.2 % intensity was used and the signals from 493 nm to 579 nm (eGFP) and 622 nm to 759 nm (FM4-64) were detected with GaAsP Detector. Pinhole size and laser dwell time was minimized. The images were recorded with time lapse mode. Image calculation, finalization and intensity measurements were carried out with Zen2012 program.

## 2.12 Inositol phosphates profile analysis

Inositol phosphates extraction was performed based on the previous protocol (Azevedo and Saiardi, 2006) with slight modifications. All of material and equipment for inositol phosphate extraction were kindly provided by Prof. Adolfo Saiardi Lab (Medical Research Council, Laboratory for Molecular Cell Biology, University College London). A qualitative analysis of inositol phosphate is divided into three general steps: labelling of *S. commune* with [$^3$H]-inositol, extraction of total soluble inositol phosphates, and inositol phosphate separation by liquid chromatography. As an alternative, higher inositol phosphate ($IP_6$-$IP_7$-$IP_8$) is also possible to detect qualitatively using polyacrylamide gel electrophoresis and visualize phosphate with Toluidine blue staining (Pisani *et al.*, 2014).

### 2.12.1 Radioactive labelling and extraction of inositol phosphates

Fully grown mycelia were homogenized in 100 ml of MM-U (see Table 1) using laboratory blender (Waring Products, Inc., Waring Laboratory, Torrington, CT, USA). The liquid cultures were diluted 1:1 in MM-U and were homogenized with laboratory blender. 3 ml of diluted cultures were labeled with 15 µl of Myo-[2-$^3$H(N)]-Inositol 1 mCi/ml (Perkin Elmer, MA, USA) for 24 h. The labeled mycelia were passed 10 times through 18G needle using a 10 ml syringe and transferred to 50 ml tube. The mycelia were pelleted at 4000 rpm for 5 min and washed with ultrapure water twice. Mycelia was transferred to 1,5 ml tube and pelleted at 14000 rpm for 5 min. Extraction of soluble inositol phosphates were performed using freshly made 500 µl extraction solution (1 M $HClO_4$, 5 mM EDTA) and 500 µl of glass beads. The mixture was vigorously disrupted using a cell disruptor (Digital Disruptor Genie- Scientific Industries, Inc., NY, USA) for 10 min and centrifuged at 4°C, 14.000 rpm for 5 min. Liquid phase containing soluble inositol phosphates was transferred to a new 1,5 ml tube. The liquid phase was neutralized using freshly made neutralization buffer (1 M $K_2CO_3$, 5 mM EDTA). The tube was incubated on ice for 2 h while the cap remained open. The tube was flipped every 15-30 min and centrifuged at 4°C, 14000 rpm for 5 min. Supernatant containing inositol phosphates was transferred to a new tube without disturbing the pellet.

### 2.12.2 HPLC run and detection using scintillation counter

The entire soluble inositol phosphates from the extraction was analyzed by Strong Anion Exchange (SAX) HPLC using SAX 4.6X125 mm column (Hichrom, UK). Sample was injected to the HPLC. HPLC elution gradient separated radiolabeled different soluble inositol phosphate. The column was eluted by mixing buffer A (1 mM $Na_2EDTA$) and Buffer B (1 mM EDTA; 1.3 M $(NH_4)_2HPO_4$, pH 3.8 with $H_3PO_4$) as following: 0-5 min, 0% buffer B; 5-10 min, 0-10% buffer B; 10-75 min, 100% buffer B; 75-85 min, 100% buffer B; 85-86 min, 0% buffer B; 86-96 min, 0% buffer B. The approximate retention times for the isomers are as follows: $IP_2$, ~16 min; $IP_3$, ~26 min; $IP_4$, ~35 min; $IP_5$, ~44 min; $IP_6$, ~58 min; $IP_7$, ~66 min; and $IP_8$, ~73 min. Four ml of Ultima-Flo AP liquid scintillation cocktail (Perkin-Elmer, MA, USA) was added to each inositol phosphates fraction. Radioactivity was quantified in a scintillation counter (Beckman Coulter,Inc., CA, USA).

## 2.13 Other molecular biology methods

### 2.13.1 Polymerase chain reaction (PCR)

DNA segment was amplified by polymerase chain reaction using specific primers and a wide range selection of DNA polymerase, depending on each primary application. Q5 and Phusion DNA Polymerase (NEB) were selected for cloning application since these two polymerases has proofreading ability that results in high accuracy for DNA amplification. Using Q5 and Phusion, the nucleotides were checked and mismatch in nucleotides are corrected during DNA synthesis. On the other hand, DreamTaq (Thermo Fisher Scientific) and OneTaq (NEB) were opted for standard application. PCR component mix contains a selected DNA polymerase, specific buffer, 10 mM dNTPs, 10 mM of each primer and template (~50 ng genomic DNA or 1-5 ng plasmid DNA) were mixed in PCR grade water (Carl Roth, Karlsruhe or 5 Prime, Gaithersburg, USA). PCR was carried out in a thermocyler T3 Thermocycler (Biometra, Göttingen), T Thermocycler (Biometra, Göttingen), Uno II (Biometra, Göttingen), or T professional thermocycler (Biometra, an Analytik Jena company, Jena) based on the manufacturers guideline. Several PCR modifications were performed for specific purpose.

(1) Touch down PCR: Touch down PCR is one of PCR modification that was used to increase specificity of PCR reactions by setting up a cycling program where the annealing temperature is gradually reduced 1-2°C every second cycle. The initial annealing temperature was set up 5-10°C above the estimated Tm of the primers.

(2) Gradient PCR: Gradient PCR was used to determine the optimum annealing temperature to solve the problem of non-specific PCR product. A gradient of annealing temperature typically 5°C below and 5°C above the expected annealing temperature, was set using the gradient function of a themocycler block T professional thermocycler.

(3) Colony PCR: To screen a high number of transformants, *E. coli* colony were touched and picked with a 10 µl pipette tip from Standard I agar plate. The colonies were used instead of DNA template in PCR reaction mix (total volume of 20 µl). A batch colony PCR was used, if there was more than 100 putative transformants obtained. Amount of 5 to 20 colonies were mixed in 20 µl PCR grade water and 1 µl of this mixture was used as template. Further PCR was carried out using a single colony if a positive band appeared in one batch colony PCR.

(4) Reverse-Transcriptase PCR (RT-PCR): RT-PCR is used to study gene expression. For this purpose, RNA molecules are converted into complementary DNA (cDNA) sequences by reverse transcriptase. cDNA was used as template instead of genomic DNA. Primer pairs were designed by including intron in a target amplicon. It is necessary to include a positive control (genomic DNA as template), the PCR product band was expected to have a higher molecular weight because of the presence of intron. RT-PCR was performed using a QuantiTect Reverse Transcriptase Kit (Qiagen). Total RNA up to 1 µg was used to synthesize 1.4 - 2 µg cDNA.

### 2.13.2 Real Time-quantitative PCR (RT-qPCR)

Appropriate set up for RT-qPCR was adapted from MIQE, Minimum Information for Publication of Quantitative Real-Time PCR Experiments Guideline (Bustin *et al.*, 2009; Taylor *et al.*, 2010). Two different RT-qPCR equipment were employed:

(1)Cepheid cycler (Cepheid, Sunnyval, USA): Each RT-qPCR reaction mix contains 12.5 µl Maxima SYBR Green/ROX qPCR Master mix (Thermo Fisher Scientific, Hamburg, Germany), 1 µl of 10 pmol/µl of each forward and reverse primer (Eurofins Genomic, Germany), 9.5 µl PCR grade water, and 2 µl of 50-100 ng/µl cDNA were transferred into a Cepheid tube. The fluorescence of SYBR Green was detected in optical channel 1 (excitation wavelength 450-495 nm, emission wavelength 510-527 nm).

(2) qTOWER$^3$ (Analytik Jena AG, Jena): A skirted white 96 well plate (VWR, Radnor, PA, USA) was used for the RT-qPCR assay. Reaction mix for RT-qPCR comprises of 3.1 µl Maxima SYBR Green/ROX qPCR Master mix (Thermo Fisher Scientific, Hamburg, Germany), 0.5 µl of 10 pmol/µl of each forward and reverse primer (Eurofins Genomic, Germany), 100 ng cDNA, and the reaction was adjusted with PCR grade water to total volume of 6.2 µl. The SYBR Green fluorescence was detected in optical channel blue (excitation wavelength 470 nm, emission wavelength 520 nm). Measurement was repeated technically three times. The RT-qPCR experiments were performed with 3 biological replicates for each mycelium and with 3 technical RT-qPCR replicates for each gene. Relative quantification of gene expression was normalized with reference genes and PCR efficiency was corrected as described in Pfaffl 2001). Reference genes used for normalization are *tef1* (JGI ID Schco3 84142) coding for translation elongation factor TEF1a, *act1* (JGI ID schco3 83632) coding for cytoskeletal protein actin, *ubi* (JGI ID schco3 71656) coding for ubiquitin conjugating enzyme.

### 2.13.3 DNA restriction endonucleases

Fragmentation of DNA for downstream analysis such as cloning was done according to different manual instruction of endonuclease products (NEB, Thermo Scientific, or Jena Bioscience). A single or double digestion of DNA was performed using two different endonucleases with a suitable, optimal buffer selection.

### 2.13.4 DNA sequencing

DNA sequencing, typically PCR amplicon or fragmented plasmid DNA, was performed by GATC Biotech (Konstanz) using either a universal primer (M13, SP6, ITS) or a primer pairs which designed specifically for product verification. Analysis of data (fasta format) was performed using Vector NTI v.11 (Thermo Fisher Scientific) and/or BLAST (NCBI).

### 2.13.5 Plasmid DNA preparation

Preparation of plasmid DNA was done rapidly using different kits depending on the amount of plasmid DNA we aim to acquire. To obtain up to 20 µg of plasmid DNA, Thermo Scientific GeneJET Plasmid Miniprep (Cat No. K0503) was used. In principle, *E. coli* cells were pelleted and dissolved in SDS-alkaline lysis to release the plasmid DNA. The lysate was neutralized to generate the optimal conditions for binding of plasmid DNA on the silica membrane in the spin column. The plasmid DNA was washed to remove contaminants and eluted with Tris-HCl buffer, pH 8.5. To acquire yield up to 500 µg, plasmid DNA was purified based on the anion-exchange resin using QIAGEN® Plasmid Maxi Kit (Cat No. 12162). The isolated plasmid DNA were used for downstream techniques such as cloning, PCR, and transformation.

### 2.13.6 Determination of Protein Kinase C activity

The PepTag® non-radioactive Protein Kinase C (PKC) determination assay (Promega GmbH) was selected to measure qualitatively PKC activity based on phosphorylation. A unique peptide PLSRTLSVAAK possessed by PKC binds to a highly specific fluorescent peptide substrate which is provided in the kit. Furthermore, phosphorylation of the peptide changes the net charge from +1 to –1, thus allowing phosphorylated and non-phosphorylated PKC to be separated on an agarose gel. The procedure for PKC assay was done according to manufacturer's protocol. Volume of 5 ml cold PKC extraction buffer was added to 1 g frozen mycelia powder. The lysate was mixed and incubated on ice for 5 min. The lysate was centrifuged at 14000 rpm, 4°C for 5 min. DEAE cellulose was pre-equilibrated using 5 ml PKC extraction buffer. Supernatant from the centrifuged lysate was aspirated and passed through the column of DEAE cellulose (Whatman® DE52, Sigma-Aldrich). PKC containing fraction in the column was eluted with an extraction buffer containing 200mM NaCl. The assay was prepared on ice bath as following: 5 µl PepTag® PKC Reaction 5X Buffer, 5 µl PepTag® C1 Peptide (0.4µg/µl), 5 µl PKC Activator 5X Solution, 1 µl Peptide Protection Solution, and 1-10 µl sample and the volume was adjusted to final volume of 25 µl using deionized water. Samples were incubated in a water bath at 30°C for 30 min. The reaction was inactivated at 95°C. Thereafter, agarose electrophoresis (0.8 % agarose in 50 mM Tris/HCl, pH 8.0) was performed to separate the phosphorylated and non-phosphorylated PKC. Gel was run at 100V for 20 minutes and visualized under ultraviolet light.

## 2.14 *In silico* analysis

### 2.14.1 Inferring phylogeny

To construct a phylogenetic tree the coding sequence information was chosen (amino acid sequence is also possible to perform). Coding sequences and amino acid sequences were obtained from NCBI. If coding sequences is selected, nucleotide sequences were translated into amino acid sequences using Transeq (http://www.cbs.dtu.dk/services/RevTrans/). Correct translated of coding sequence to amino acid were visualized using Launch Jalview Desktop (http://www.jalview.org/) and expected length of amino acid was ensured. Coding region sequences were aligned with MAFFT using G-INS-I which was implemented in Jalview (web service > alignment > run MAFFT with preset). Different preset is sometimes worthwile to try in order to create the best alignment quality, depending on the input sequences. Online tool RevTrans (http://www.cbs.dtu.dk/services/RevTrans/) was used to reverse translate the amino acid alignment into a codon alignment. Alignment was loaded back in Jalview and alignment parts that are not considered for calculation were noted somewhere in a word document. Tree calculation was done with MrBayes after the alignment file was converted from FASTA to NEXUS format using ALTER (https://sing.ei.uvigo.es/ALTER/). The phylogeny reconstruction with MrBayes on XSEDE (3.2.6) was carried out via the online tool CIPRES Science Gateway (https://www.phylo.org/portal2/login!input.action). The codon alignment in NEXUS file format was uploaded to the main task folder. The obtained tree file (.tre) was visualized using FigTree v1.4.3 and converted to a Portable Network Graphics file (.png). If necessary, tree captions and information were edited using Adobe Illustrator.

### 2.14.2 Chromosomal organization of *imp* surrounding gene

For the analyses of the *imp* chromosomal organization of *S. commune* H4-8, TatD and LoeD the JGI based tools available at the genome homepages were utilized. The distances between genes were searched, locations and functions were added manually into a Windows Editor (.txt file). The software ChromoMapper (Niculita-Hirzel, 2008) was utilized to display the imp gene surrounding then later all strains were compared to the reference strain H4-8.

### 2.14.3 Other protein analysis

#### 2.14.3.1 Motif Analysis and Protein structure modelling

Powerful tool Multiple Em for Motif Elicitation (MEME) was chosen to define the motif regions for inositol monophosphatase orthologues. The online server I-TASSER was selected to obtain the protein 3D structure modelling (Roy *et al.*, 2010; Yang *et al.*, 2015; Zhang J. *et al.*, 2015; Zhang, 2008). Protein sequences were obtained either from JGI or NCBI. The default setting was left out as it is.

# 3 Results

## 3.1 Inositol phosphate in *Schizophyllum commune*

Inositol phosphate is conserved among eukaryotes, including the basidiomycete *S. commune*. Inositol and different inositol phosphate species, from inositol monophosphate to several inositol polyphosphates, were successfully detected using Strong Anion Exchange-High Liquid Performance Chromatoraphy (SAX-HPLC) in *S. commune* (Figure 10).

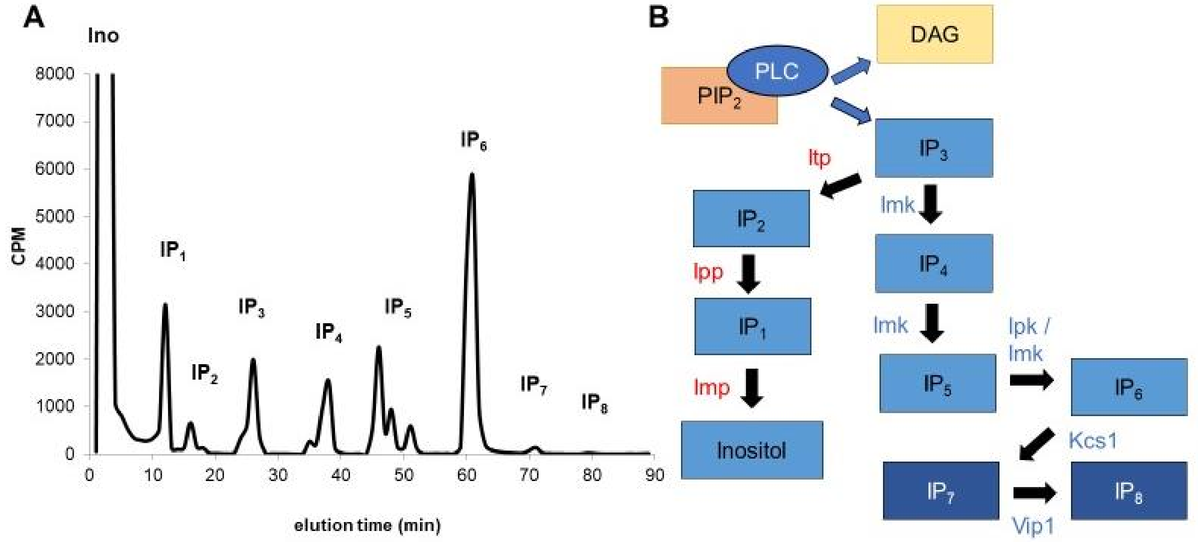


Figure 10. SAX-HPLC elution of inositol phosphates in *S. commune* (A). Simplified pathway of inositol phosphate metabolism in *S. commune* shows the inositol phosphates phosphatases (written in red) and inositol phosphate kinases (written in blue) are obviously present in *S. commune* (B). $PIP_2$: phosphatidylinositol 4,5 bisphosphate; DAG: diacylglycerol; PLC: phospholipase C; $IP_1$: inositol monophosphate; $IP_2$: inositol bisphosphate; $IP_3$: inositol trisphosphate; $IP_4$: inositol tetrakisphosphate; $IP_5$: inositol pentakisphosphate; $IP_6$: inositol hexakisphosphate; $IP_7$: diphosphoinositol-pentakisphosphate; IP8: bis-diphosphoinositol-tetrakisphosphate; Itp: inositol trisphosphate phosphatase; Ipp: inositol polyphosphate phosphatase; Imp: inositol monophosphate phosphatase/ inositol monophosphatase; Imk: inositol polyphosphate multikinase; Ipk: inositol 2-pentakisphosphate kinase; Kcs1: inositol polyphosphate kinase/ inositol-hexakisphosphate 5-kinase; Vip1: Inositol polyphosphate kinase 2.

Inositol trisphosphate ($IP_3$) is the center of inositol phosphate metabolism, derived from the cleavage of phosphatidylinositol bisphosphate ($PIP_2$) by a phosphoinositide-phospholipase C (PI-PLC/PLC). Inositol monophosphate ($IP_1$) and inositol bisphosphate ($IP_2$) are the intermediate precursors for inositol production and both molecules are the result of dephosphorylation product from the prominent second messenger molecule $IP_3$. In *S. commune*, inositol polyphosphates species (inositol tetrakisphosphate/ $IP_4$, inositol pentakisphosphate/ $IP_5$ and inositol hexakisphosphate/ $IP_6$) are promptly generated from molecule $IP_3$, suggesting the existence of inositol phosphate kinases in this filamentous fungus. As it is clearly shown in the SAX-HPLC elution profile, fungal $IP_6$ can be further phosphorylated to diphosphoinositol-pentakisphosphate ($IP_7$) and bis-diphosphoinositol-tetrakisphosphate ($IP_8$). $IP_7$ and $IP_8$ were classified as the high energy signaling molecules termed as inositol pyrophosphates. The analysis clearly indicates that inositol pyrophosphates are obviously conserved in *S. commune*. Inositol and inositol phosphate have been known as mediator for growth and sexual reproduction in ascomycete fungi. Inositol polyphosphate, for instance, has been known to promote fruiting body maturation and controls the amount of developed fruiting bodies after fertilization in ascomycete (Xie *et al.*, 2017). In this study, SAX-HPLC analysis after inositol phosphate extraction from different developmental stage of *S. commune* was performed. As it is shown in the Figure 10, fruiting bodies of *S. commune* produces relatively low inositol hexakisphosphate (IP$_6$) level, thus there is no further generation of inositol pyrophosphates (IP$_7$ and IP$_8$). This result implies that IP$_6$ and further phosphorylated high energy inositol pyrophosphates are not necessarily needed during fruiting body maturation in *S. commune*, which is quite the opposite from the evidence that is shown in ascomycetes. Furthermore, fruiting bodies lack the production of IP$_{5a}$ which is presumably identified as Inositol 1,3,4,5,6-pentakisposphate. Inositol 1,3,4,5,6-pentakisposphate is known to be the precursor for generating the full phosphorylated inositol phosphate IP$_6$, thus it makes sense that the production of IP$_6$ seems to be low. On the other hands, IP$_{5b}$ and IP$_{5c}$ are the dephosphorylation product generated from IP$_6$. IP$_6$ is the most abundant inositol polyphosphate in the basidiomycete fungus *S. commune*.

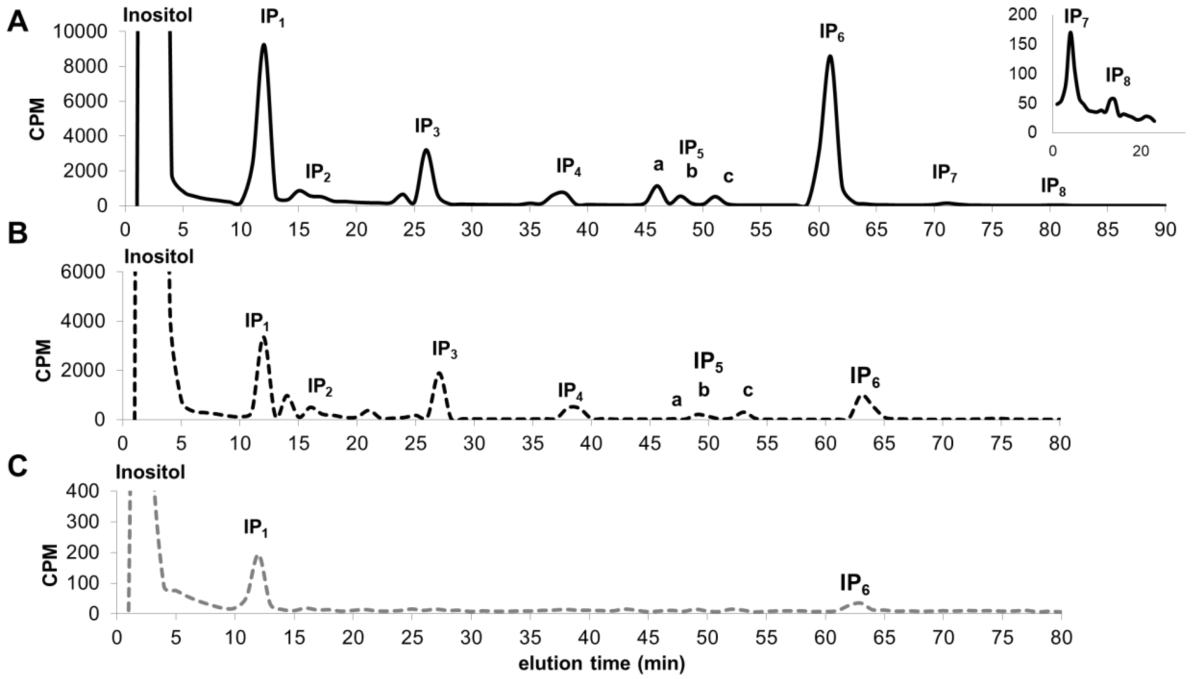


Figure 11. SAX-HPLC analysis of inositol phosphate in different sexual developmental stages. Mycelia from wild type 4-39 (A), fruiting bodies from wild type 12-43 x 4-39 (B), and spores from wild type 12-43 x 4-39. In fruiting bodies and spores, the elution is limited only until 80 min, considering there will be no inositol pyrophosphates produced from very small amount of IP$_6$.

In Figure 11, it is shown that IP$_6$ is the most prominent inositol polyphosphate in three different developmental stages (monokaryon, fruiting bodies, and spore). Inositol phosphate profile in basidiospores collected from a petri dish culture was analyzed as well using SAX-HPLC. Interestingly, we could see similar inositol phosphate profile in spores and fruiting bodies. The spores lack of inositol polyphosphates phosphorylated from $IP_3$, however they still have relatively great amount of $IP_1$. The starting material of basidiospores used for inositol phosphate extraction was very low amount (0.5 mg), therefore it reflects the low inositol phosphates level. Lower species of inositol phosphate namely $IP_2$ and $IP_1$ are the phosphorylated products from the prominent molecule $IP_3$ which are found abundant in fruiting bodies. This fact suggests that these two molecules are important precursors for inositol generation, in line with the result from the ascomycete (Xie *et al.*, 2017) that pinpoint inositol as essential substance for fruiting body maturation.

## 3.2 Inositol phosphate phosphatase and inositol phosphate kinases

*In silico* analysis of genes involved in the interconversion of inositol phosphate in *S. commune* was performed in this study. *S. commune* possesses several inositol phosphate phosphatases and inositol phosphate kinases (Table 11). Comparative analysis was performed with baker yeast *S. cerevisiae* that has been studied intensively as a model organism to study inositol phosphate signaling for more than 20 years (Culbertson *et al.*, 1976).

**Table 11. Proteins involved in inositol phosphate metabolism in *S. cerevisiae* 288c and their homologues in *S. commune* H4-8**

| No. | *S. cerevisiae* Protein | NCBI Protein ID *S. cerevisiae* | *S. commune* protein homologues | | | | Information |
|---|---|---|---|---|---|---|---|
| | | | schco1 | Protein Identity via BLAST NCBI | schco3 | Protein Identity via BLAST JGI | |
| 1 | Inositol polyphosphate kinase Kcs1/ Inositol-hexakisphosphate 5-kinase | NP_010300 | 46826 | Score 628<br>E-Value 1.05e-053<br>Identity 58.7%<br>Subject coverage 44.8% | 2609005 | Score 736<br>E-Value 6.75e-056<br>Identity 58.7%<br>Subject coverage 19.6% | Phosphorylation from Ins 1,2,3,4,5,6-$P_6$ to 5-PP-$IP_5$<br>Phosphorylation from 1-PP-$IP_5$ to $[PP]_2$-$IP_4$ |
| 2 | Phosphatidylinositol phospholipase C, delta | NP_015055 | 79892 | Score 851<br>E-Value 3.67e-061<br>Identity 41.6%<br>Subject coverage 49.2% | 2645684 | Score 851<br>E-Value 2.11e-060<br>Identity 41.6%<br>Subject coverage 41.0% | PLC catalyzes the hydrolysis of membrane PIns-$P_2$, generating the second messengers Ins 1,4,5-$P_3$ and DAG |
| | | | 82573 | Score 847<br>E-Value 2.16e-069<br>Identity 37.4%<br>Subject coverage 50.8% | 2628010 | Score 847<br>E-Value 2.43e-069<br>Identity 37.4%<br>Subject coverage 50.8% | |
| | | | 109536 | Score 370<br>E-Value 8.17e-021<br>Identity 41.8%<br>Subject coverage 28.9% | 2669046 | Score 428<br>E-Value 2.04e-028<br>Identity 45.0%<br>Subject coverage 31.1% | |
| 3 | Phosphoinositide 5-phosphatase Inp54/ inositol-1,4,5-trisphosphate 5-phosphatase | NP_014576 | 81429 | Score 299<br>E-Value 1.04e-013<br>Identity 33.7%<br>Subject coverage 42.6% | 2661211 | Score 299<br>E-Value 1.27e-013<br>Identity 33.7%<br>Subject coverage 41.7% | Dephosphorylation from Ins 1,3,4,5-$P_4$ to Ins 1,3,4-$P_3$<br>Dephosphorylation from Ins 1,4,5-$P_4$ to Ins 1,4-$P_2$ |

| | | | | | | | |
|---|---|---|---|---|---|---|---|
| 4 | inositol polyphosphate multikinase | NP_010458 | 70406 | Score 436<br>E-Value 2.51e-031<br>Identity 44.1<br>Subject coverage 55.0% | 2639210 | Score 436<br>E-Value 2.89e-031<br>Identity 44.1<br>Subject coverage 55.0% | Phosphorylation from Ins 1,3,4,5-$P_4$ to Ins 1,3,4,5,6-$P_5$<br>Phosphorylation from Ins 1,4,5-$P_3$ to Ins 1,4,5,6-$P_4$<br>Phosphorylation from Ins 1,4,5,6-$P_4$ to Ins 1,3,4,5,6-$P_5$<br>Phosphorylation from Ins 1,3,4,6-$P_4$ to Ins 1,3,4,5,6-$P_5$ |
| 5 | Inositol polyphosphate kinase, Vip1 | NP_013514.1 | 63048 | Score 2140<br>E-value 0.0E000<br>Identity 56.1<br>Subject coverage 77.7 | 2621615 | Score 2157<br>E-value 0.0E000<br>Identity 56.1<br>Subject coverage 70.4 | Bifunctional inositol kinase that acts in concert with the IP6K kinase KCS1 to synthesize the diphosphate group-containing inositol pyrophosphates diphosphoinositol pentakisphosphate, PP-InsP5, and bis-diphosphoinositol tetrakisphosphate, (PP)2-InsP4 |
| 6 | Inositol pentakisphosphate 2-kinase | NP_010601 | - | - | - | - | Phosphorylation from Ins 1,3,4,5,6 $P_5$ to Ins 1,2,3,4,5,6-$P_6$ |
| | *homologue with human* | | 51281 | NCBI Blast<br>Max score 82.4<br>Total score 82.4<br>Query cover 35%<br>E-value 4e-17<br>Identity 31% | 2608225 | Score 230<br>E-Value 3.75e-008<br>Identity 48.9%<br>Subject coverage 17.3% | Phosphorylation from Ins 1,3,4,5,6 $P_5$ to Ins 1,2,3,4,5,6-$P_6$ |
| 7 | Inositol monophosphate 1-phosphatase Inm2/ inositol-1(or 4)-monophosphatase | NP_010573 | 47747 | Score 502<br>E-Value 2.19e-050<br>Identity 53.0<br>Subject coverage 49.7 | 2697390 | Score 502<br>E-Value 2.19e-050<br>Identity 53.0<br>Subject coverage 49.7 | Dephosphorylation from Ins 4-P or 1-P to inositol |
| | Inositol monophosphate 1-phosphatase Inm1/ inositol-1(or 4)-monophosphatase | NP_011912 | 47747 | Score 392<br>E-Value 3.89e-035<br>Identity 50.4<br>Subject coverage 42.1 | 2697390 | Score 392<br>E-Value 4.47e-035<br>Identity 50.4<br>Subject coverage 42.1 | Dephosphorylation from Ins 4-P or 1-P to inositol |

*In silico* analysis revealed inositol phosphate metabolism in *S. commune*. Inositol phosphate metabolism employs a host of kinases and phosphatases that generate and degrade abundant signaling molecules of inositol phosphate. Set of kinases and phosphatases can be more broadened up in terms of phosphatidylinositol signaling, however for this study, only kinases and phosphatases of water-soluble inositol phosphate metabolism which were observed. A further study is needed to complete the huge network of the membrane associated phosphatidylinositol signaling cascade. Inositol pentakisphosphate 2-kinase (Ipk) that participate in the phosphorylation of inositol hexakisphosphate ($IP_6$), is absent in the yeast, but not in *S. commune* (homologue of Ipk was searched against human Ipk, see Table S1). This evidence suggests that the functional module of Ipk is possibly taken over by inositol phosphate multikinase (Imk) in *S. cerevisiae*. It has been shown that three homologs of yeast phosphatidylinositol phospholipase C (PI-PLC) has been shown in *S. commune*. As it was mentioned before, the birth of inositol phosphates highly depends on the cleavage of $PIP_2$ by PI-PLC. Once PI-PLC cleaves $PIP_2$, DAG resides in membrane and the prominent messenger molecule $IP_3$ is liberated into cytosol, which later is phosphorylated to generate inositol by various phosphatases, or it can be further phosphorylated by kinases to $IP_6$ and the energy-rich signaling molecule inositol pyrophosphate. The delineation of phosphatases is still far from complete. From what is known so far, inositol phosphate phosphatases are classified into 3 different groups (5- phosphatase, 1-phosphatase, and 4-phosphatase) but based on *in silico* analysis, only inositol-1,4,5-trisphosphate 5-phosphatase and inositol monophosphate 1-phosphatase (inositol monophosphatase) could be found in both model fungi *S. commune* and *S. cerevisiae*. Additionally, only one inositol monophosphatase homologs could be discovered in *S. commune*. Altogether Ipk, Imk, Kcs1, and Vip1 belong to the inositol phosphate kinase family, indicated by the existence of conserved motif in the inositol binding region, was found in *S. commune*.

### 3.3 Inositol phosphate kinases gene expression

As it is shown previously at section 3.1, inositol pentakisphosphate 2-kinase (*ipk*) and inositol polyphosphate multikinase (*imk*) gene expression are expected to be down-regulated in the fruiting body formation, since the production of $IP_6$ is reduced. Therefore, gene expression of *ipk* and *imk* was further analyzed in different developmental stage. Ipk and Imk are the kinases that presumably generate the inositol hexakisphosphate ($IP_6$) in *S. commune*. Unfortunately, as a result, it was shown that gene expression of *ipk* was not significantly altered in dikaryon, fruiting body and fruiting body & mycelia (Figure 12).

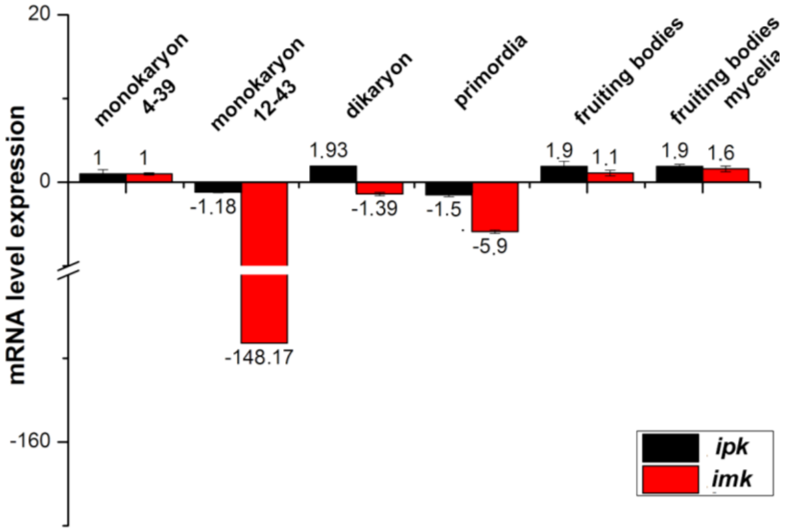


Figure 12. Gene expression of inositol pentakisphosphate 2-kinase (*ipk*) and inositol polyphosphate multikinase (*imk*) in different sexual developmental stages.

On the other hand, gene expression of *imk* was insignificantly stable in dikaryon and was significantly down-regulated in primordia within 5.9-folds. Interestingly, *imk* gene expression was stable in fruiting bodies and in fruiting bodies & mycelia. This finding is not in line with the inositol phosphate profile in different sexual developmental stages of *S. commune* which can be seen at section 3.1, in which it has been shown that $IP_6$ level is depleted. This result remains ambiguous, however the primordia which is the early stage of fruiting body shows repression of *imk* gene expression. The remarkable down-regulation of *imk* mRNA level expression in monokaryon 12-43 compared to 4-39 indicates that there is a variation of *imk* gene expression among wild-type.

### 3.4 Proteins involved in the development of *S. commune*

Protein fraction were isolated from the *S. commune* in different development stage. The abundance of proteins in dikaryon, primordia, and fruiting body was compared to monokaryon 12-43 and 4-39 respectively. Low and high abundant proteins in every developmental stages are classified based on EuKaryotic Orthologous Groups/ KOG (Figure 13 and 14).

Phosphatidylinositol-4-phosphate 5-kinase was found to be repressed in dikaryon vs. monokaryon 12-43 within 2,45-folds and in primordia vs. monokaryon 12-43 within 2,48-folds. On the other hand, phosphatidylinositol 3- and 4-kinase was repressed in fruiting body in comparison to monokaryon 12x43 within 2,10-folds and to monokaryon 4-39 within 2,42-folds. Phosphatidylinositol-4-phosphate 5-kinase generates phosphatidylinositol-4,5-bisphosphatase from phosphatidylinositol-4-phosphatase and phosphatidylinositol 3- and 4-kinase generates phosphatidylinositol-4-phosphatase and phosphatidylinositol-3-phosphatase from phosphatidylinositol. This evidence shows that phosphatidylinositol signaling is involved in the sexual development of *S. commune* and the phosphoinositides might act as negative regulators for fruiting body development.

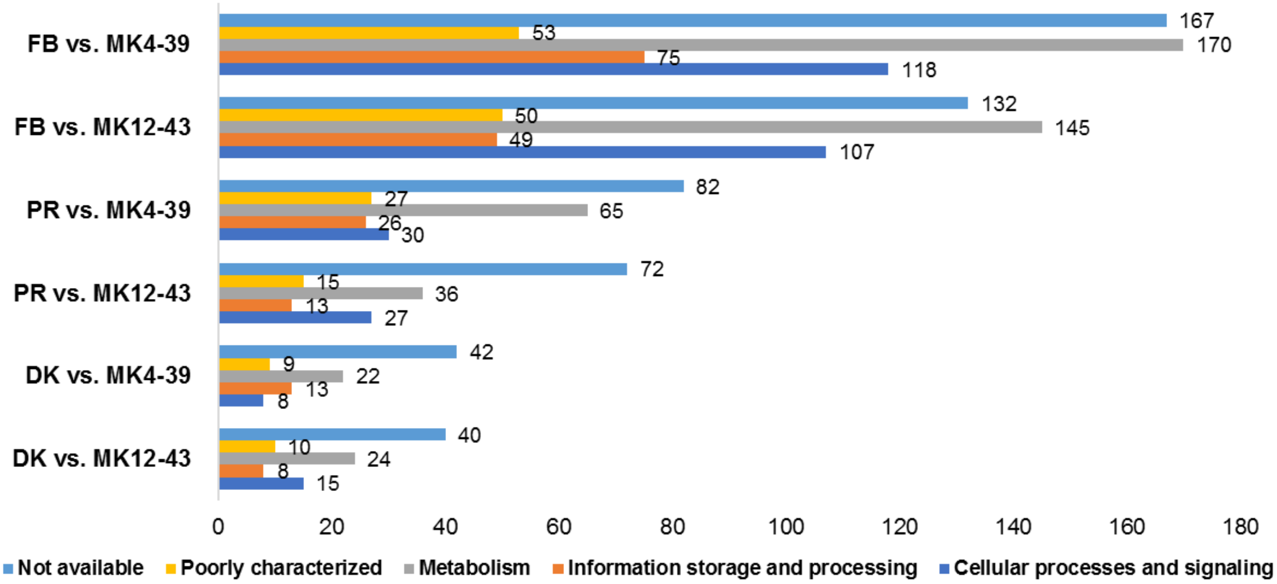


Figure 13. Classification of low abundant signal proteins based on EuKaryotic Orthologous Groups (KOG). MK: monokaryon, DK: dikaryon, PR: primordia, FB: fruiting bodies.

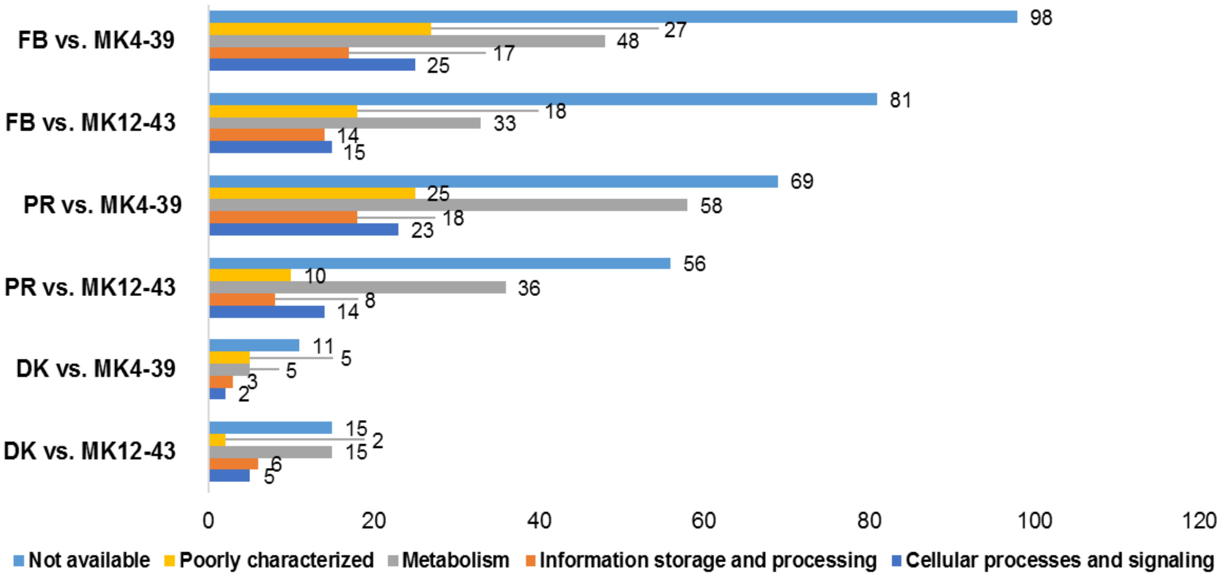


Figure 14. Classification of high abundant signal proteins based on EuKaryotic Orthologous Groups (KOG). MK: monokaryon, DK: dikaryon, PR: primordia, FB: fruiting bodies.

Repression of phosphatidylinositol signaling might affect calcium signaling which was reflected on the repression of calcium-binding EF-hand protein. Calcium-binding EF-hand protein which requires activation from $IP_3$ was found to be down-regulated as well in dikaryon and in primordia. Multicopper oxidase (schco3 Protein ID 2179879) and peptidase M35 were found to be induced in dikaryon compared to both monokaryon 12-43 and 4-39, showing that at least one of the multicopper oxidase is required for establishing dikaryon. Signal transduction response regulator was repressed in dikaryon in comparison to both monokaryons. This protein has a function in detecting and responding some changes in surrounding environment, including stressors. Interestingly, polysaccharide deacetylase that has been known to play a crucial role in the alteration of cell shape, was induced in primordia and fruiting bodies. As it has been known that in primordia and fruiting body, hyphae alter its structure to become thicker hyphae since the dikaryotic hyphae form aggregates.

Protein that possesses histidine acid phosphatases (HAP) domain was repressed in the fruiting bodies and low abundant of putative inositol phosphatase was observed also in the fruiting bodies. In line with the repression of lipid inositol phosphate kinase, it is found that myo-inositol-1-phosphate synthase was also repressed only in the fruiting bodies compared to monokaryon 12-43. The repression of myo-inositol-1-phosphate synthase indicates that the production of inositol is reduced and therefore, limiting also the lipid inositol phosphate (phosphatidylinositol/ PI) which was reflected on the repression of phosphatidylinositol 3- and 4-kinase in fruiting bodies. Several ATPases were also repressed in fruiting body, in agreement with the repression of HAP domain which is responsible for inositol pyrophosphate generation. Inositol pyrophosphates are capable to regulate many biological processes most probably by controlling energetic metabolism and ATP production. Less abundant of ATPase in the proteome seems to be an effect from the low abundance of inositol pyrophosphate in fruiting body or *vice versa*.

Multicopper oxidase (schco3 Protein ID 2179879) as well as glycosyltransferases were induced in primordia formation. Glycosyltransferases are another large multigene family in eukaryote that responsible for biosynthesis of secondary metabolites, and sugar conjugation results in increased stability and water solubility.

Aldehyde dehydrogenase and spermine synthase were induced in primordia. On the other hand, beta tubulin and actinin was repressed in the primordia within 2,03-folds and 2,63-folds, respectively. Glycoside hydrolase family were repressed in primordia but elevated in fruiting bodies, as fungal glycoside hydrolases (GH) is known to act on cell wall components and are responsible for morphological changes, possibly from primordia to fruiting body. Furthermore, several GMC oxidoreductases, peptidases, protein kinases, and ribosomal protein family were also repressed in primordia.

According to the proteome analysis, there are 161 and 215 proteins that are induced in fruiting bodies compared to monokaryon 12-43 and 4-39, respectively. In other basidiomycetes, cytochrome P450 is induced in fruiting body, indicating that this oxidative enzyme might play a role in mushroom development. Moreover, several proteins that are important in carbohydrate metabolism are elevated in fruiting body including glycolysis transferase (GT) family, glyoxalase I, glycoside hydrolase (GH) family, polysaccharide deacetylase, and esterase/ lipase/ thiosterase. Metallophosphoesterase that is required for GPI-anchor proteins transport from the organelle endoplasmic reticulum, found to be induced and repressed in fruiting bodies.

There are 483 and 583 proteins that are repressed in the fruiting body stage compared to monokaryon 12-43 and 4-39, respectively. Mostly repressed proteins are metabolism associated proteins and cellular processes and signaling associated proteins (Table S11-S22). Several microtubules and microfilaments associated proteins were repressed in fruiting body stage, presumably the growth of hyphae is only required in the mycelial expansion but not necessarily needed in fruiting bodies. In fruiting bodies, several proteins that are involved in motor activity such as actin, actin-binding, Arp2/3 complex, alpha tubulin, beta tubulin, F-actin capping protein, kinesin, and myosin, were found lower in abundance. Additionally, dynamin, importin-beta, NSF attachment protein, clathrin propeller, and clathrin adaptor complex were also repressed in fruiting bodies (Table S21 and S22). This evidence shows that the transport activity in fruiting bodies is deactivated. Further, several proteins related to signaling such as Ras small GTPase family, GTP-binding nuclear protein Ran, calcium-binding EF-hand, regulator of G-protein signaling, fungal G-protein alpha subunit, and signal transduction histidine kinase, were repressed as well in fruiting bodies.

## 3.5 Lithium effect on inositol phosphate in *S. commune*

### 3.5.1 Lithium effect on the growth and mating in *S. commune*

The lithium induces growth inhibition of fungus was tested in relevant concentration (5 mM lithium chloride). The lithium-treated fungus showed peculiar and irregular fungal colony shape accompanied with the significant growth reduction (Figure 15). The growth of wild type was inhibited fifty five percent by lithium. Occasionally, multi-branching and retarded hyphae was observed under microscopy, however it is not concluded as a specific effect of lithium. Meanwhile growth and hyphal morphology is not altered in the fungus treated with 5 mM KCl as control. This evidence indicates strongly that the growth inhibition is solely caused by lithium.

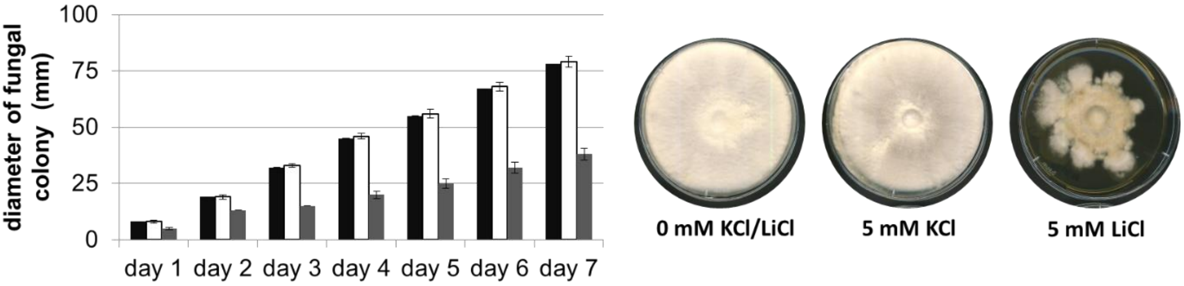


Figure 15. Growth rate of lithium treated wild-type 4-39 compare to control fungus growth on CYM without KCl/LiCl and control with 5 mM KCl. Black bar: 0 mM KCl/LiCl, white bar: 5 mM KCl, and grey bar: 5 mM LiCl.

Higher concentration of lithium was applied to the fungal culture with much greater effect on growth inhibition. Starting from 15 mM LiCl concentration, the wild-type failed to grow. The growth of lithium-treated mating culture was slightly reduced, and brown pigment is excreted into the solid media, suggesting a stress response towards lithium supplementation (Figure 16 F-G). In mating plate without LiCl addition, the aggregation of dikaryon mycelia (so-called primordia) started to develop in 10-day post inoculation (dpi) and after 13 days cultivation, the primordia developed to fully mature fruiting bodies that bear basidiospores which able to germinate to the haploid monokaryon.

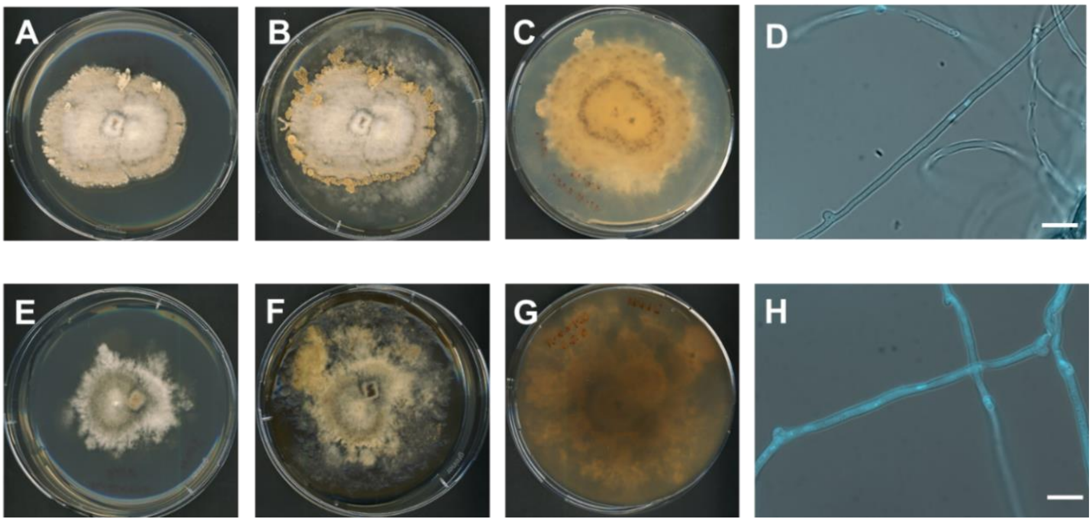


Figure 16. Lithium affects mating in *S. commune*. A-D represent a wild type 12-43 x 4-39 without lithium presence (5 mM LiCl), E-H represent a lithium-treated 12-43 x 4-39. Mycelia of mating culture 12-43 x 4-39 were documented after 13 days (A, E) and 25 days post inoculation (B-C, F-G). A brown pigment is visible only in the lithium-treated mating culture (G) and is absent in the control mating plate (C). Bar size is 10 µm.

In contrast to the non-treated fungi, which possessed thick aerial mycelia, the lithium treated fungi, on the other hand showed an unusual ring formation in the middle of the fungal colony and irregular colony shaped (Figure 16 E and F) as it was shown in the haploid monokaryon treated with lithium. Less aerial submerge mycelia were observed in the lithium treated mating culture. Aggregation of mycelia was observed only in some section and the mating culture failed to develop any primordia or fruiting body, although the clamp connection was established. Binucleated cell was occasionally observed in the hyphae, even though it is hard to observe due to formation of many vesicles or unidentified inclusion bodies along the hyphae.

### 3.5.2 Lithium effect on inositol phosphate and inositol monophosphatase activity

Several investigations provide evidence that lithium mediates the inhibition of inositol monophosphatase and other member of phosphatases, affecting inositol phosphate metabolism. Measurement of inositol and inositol phosphates levels in lithium-treated wild type *S. commune* 4-39 were performed using a SAX-HPLC. The evidence shows that lithium affects the entire metabolism of inositol phosphate. It is expected that the inositol level is decreased in the presence of lithium, however the evidence shows insignificant reduction in inositol level. The insignificant reduction of inositol was perhaps due to the trouble on washing step before inositol extraction, therefore the inositol is not shown in Figure 17, however the ratio of inositol between the treated and untreated wild type *S. commune* was calculated and shown in Figure 18.

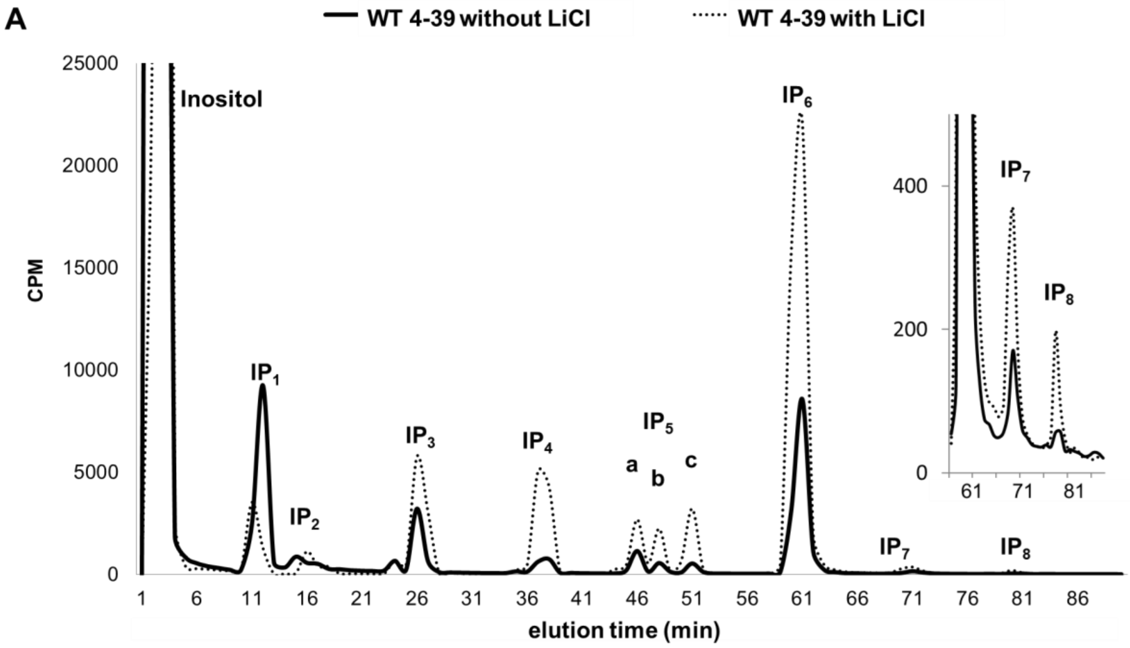


Figure 17. HPLC elution profile of soluble inositol phosphates of *S. commun*e wild type 4-39. Mycelia were labeled 24 h in the absence (black line) or in the presence of lithium (black dotted line) before extraction and SAX-HPLC analysis. The chromatogram of inositol pyrophosphate $IP_7$ and $IP_8$ was represented in a different scale. $IP_{5a}$ species in *S. commune* was identified as inositol 1,3,4,5,6-pentakisphosphate. The Inositol phosphate profile shows in this figure is a representative result from three times measurement.

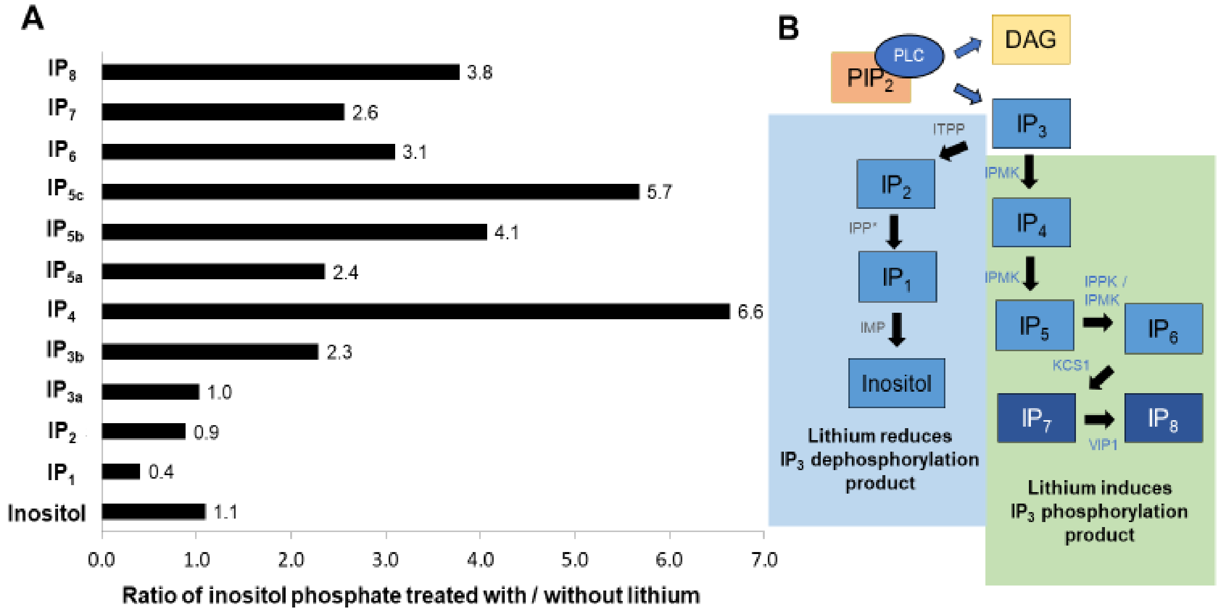


Figure 18. A. Ratio of inositol phosphate between treated and untreated lithium in wild type *S. commune* 4-39. Phosphorylated products of IP$_3$ (IP$_4$ to IP$_8$) are increased in lithium manner, suggesting the unknown mechanism of lithium's action on kinases. B. Lithium action on IP$_3$ dephosphorylation (light blue box) and phosphorylation product (green blue box).

Looking closer into the dephosphorylated product of IP$_3$ which generates inositol as the final product of dephosphorylation, almost the entire phosphorylated products were induced by lithium, except inositol monophosphate (IP$_1$) and inositol bisphosphate (IP$_2$). Inositol precursor IP$_1$ is expected to be elevated in the presence of lithium, however a conflicting indication was observed. IP$_1$ level is remarkably reduced up to sixty percent, on the other hand the IP$_2$ was reduced ten percent. This evidence should reflect a high inositol level reduction, but the result shows only a slightly decrease of inositol content.

It is indeed noticeable that the IP$_3$ molecule is accumulated in the lithium treated fungus. This implies that lithium might inhibit also the enzyme that is responsible to dephosphorylate IP$_3$ to IP$_2$ and IP$_1$ and one of them could be the 5-phosphatase enzyme (inositol 1,4,5-trisphosphate 5-phosphatase). On the other hand, lithium clearly induces higher inositol phosphate species (IP$_4$, IP$_5$, IP$_6$) including the high energy inositol pyrophosphates (IP$_7$ and IP$_8$) suggesting that the action of lithium does not only affect inositol phosphate phosphatases, but also the inositol phosphate kinases. Based on the ratio calculation between the lithium treated and non-treated wild type 4-39, the IP$_4$ precursors is the most elevated inositol phosphates. The influence of lithium on inositol phosphate profile triggers us to examine the effect of lithium on the inositol monophosphatase enzymatic activity on wild type strains 4-39. Lithium reduces inositol monophosphatase activity to sixty percent in the wild type 4-39, it is in line with the result that inositol is slightly repressed in the SAX-HPLC elution profile.

### 3.5.3 Lithium effect on *S. commune* metabolism and cellular signaling

To identify pathways and/or proteins target regulated upon lithium treatment, 2D-PAGE was performed and followed by mass spectrometry analyses. Cytosolic proteins were isolated from the wild type 4-39 and the constitutively active Ras1 mutant *ras1*$^{G12V}$. Both were cultivated with and without lithium. Proteins with change in abundance were identified by MS and functional assignment (Table S2 and S3) was done by using the KOG (EuKaryotic Orthologous Groups) classification. Induced and repressed proteins were classified into the KOG group of metabolisms, cellular processes & signaling, and information storage & processing. This classification is in remarkable agreement with the properties recently attributed to the inositol phosphates' role. The highly phosphorylated forms heavily induced by lithium, such as $IP_6$ and the inositol pyrophosphates $IP_7$ and $IP_8$ have been described as 'the metabolic messenger' working at the interface between signaling and metabolism (Wilson *et al.*, 2013). In the wild type, 68 proteins were repressed and mostly are related to metabolic function; meanwhile only 9 proteins were induced in response to the lithium treatment (Figure 19).

Interestingly, a putative ATPase F1/V1/A1 (JGI ID 1088750) was repressed only in the wild type under lithium treatment. The ATPase F1/V1/A1 is putatively responsible for hydrogen-transporting ATPase activity and catalyzing the process of transmembrane movement of substances. This evidence is in agreement with the previous study that inositol depletion is responsible for vacuolar defect and the decrease of vacuolar ATPase (Deranieh *et al.*, 2015).

In the active Ras1 mutant *ras1*$^{G12V}$, proteomic analysis revealed 26 repressed proteins and induced 29 proteins in lithium treated *ras1*$^{G12V}$ (Figure 19). The compositions of repressed and induced proteins in response to lithium are indeed different in the wild type 4-39 and *ras1*$^{G12V}$ mutant, in agreement with the striking different effect that lithium elicits on the inositol phosphate metabolism. Interestingly, expression of a protein possessed histidine acid phosphatase (HAP) domain was repressed in the wild type 4-39 (JGI ID 1135584) but induced in the constitutively active Ras1 (JGI ID 2643503) mutant. Phytase, the enzyme that catalyzes the hydrolysis of $IP_6$ to inositol, possesses HAP domain (Mullaney and Ullah, 2005; Wyss *et al.*, 1999). Repression of HAP occurs only in the wild-type is correlated with the induction of high phosphorylated inositol phosphate synthesis executed by lithium.

Furthermore, it was found that fructose 1,6-bisphosphate aldolase (ketose-bisphosphate aldolase) and fructose-1,6-bisphosphatase were repressed by lithium in the wild type and active Ras1 mutant, respectively. Both enzymes are catalysts for the intermediates of fructose-6-phosphate and as it has been known that fructose-1,6-bisphosphatase belongs to the metal-dependent and lithium-sensitive phosphomonoesterase enzyme, together with inositol monophosphatase and inositol polyphosphate 1-phosphatase (York *et al.*, 1995). Surprisingly, it was found that cytoskeleton protein actin-1 (JGI ID 1194206 and JGI ID 2645649, in the wild-type and Ras1 active mutant, respectively) was repressed in the lithium treatment on both strains. This implies that lithium may have some effect on actin dynamics, considering the importance of actin-1 in the hyphal polarization growth.

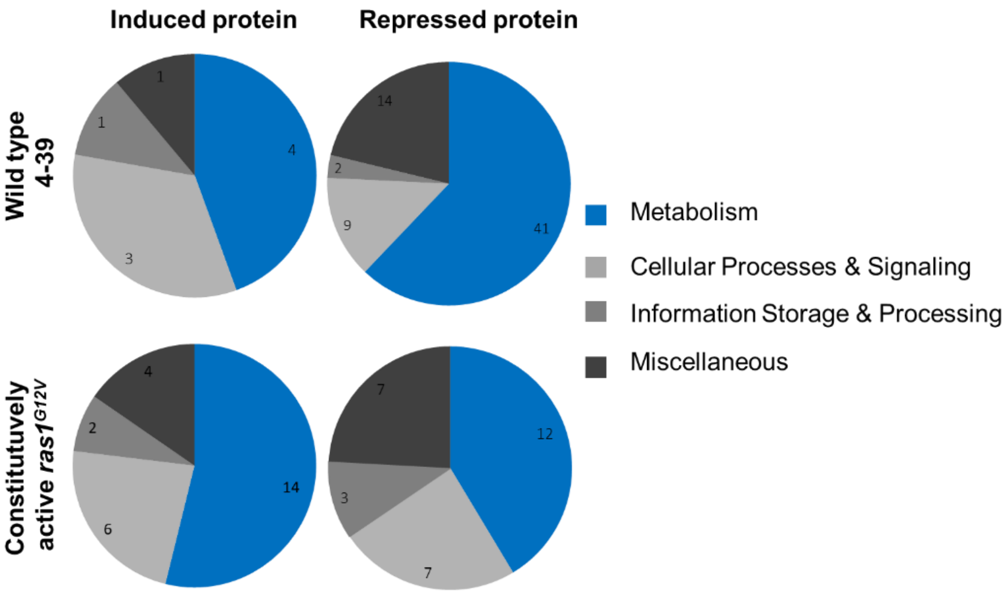


Figure 19. KOG classification of regulated proteins in *S. commune* wild type 4-39 and constitutively active Ras1 mutant *ras1*$^{G12V}$ in the presence of lithium. Lithium affects a wide range of cellular processes in *S. commune* from the metabolism, cellular signaling and Information storage and processing.

### 3.5.4 Lithium effect on actin dynamics

The evidence of low abundance of actin in proteomics level triggers the investigation of actin dynamics in *S. commune* cultivated in the presence of 5 mM LiCl.

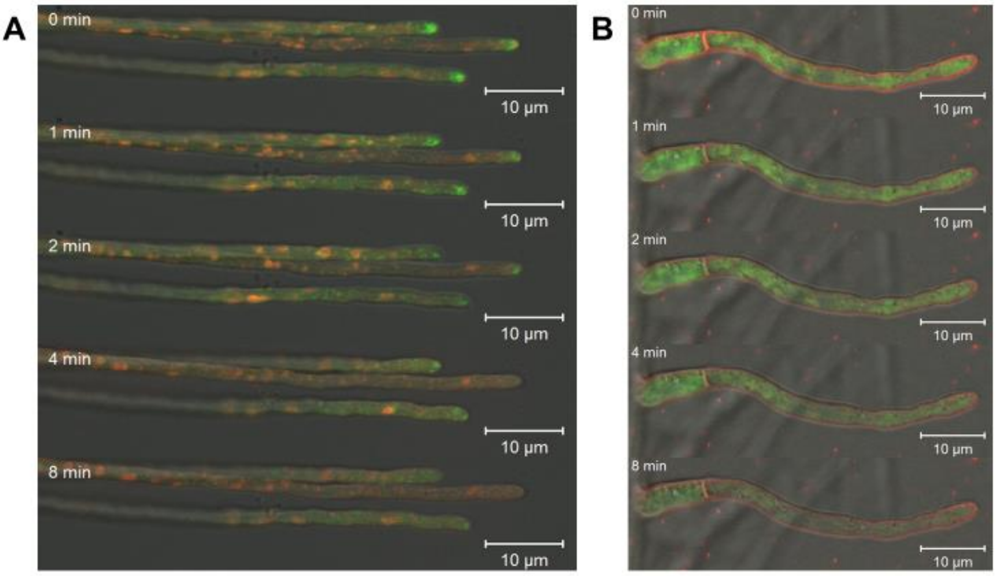


Figure 20. Actin visualization with and without lithium effect. High actin dynamics in *S. commune* using Life-act GFP without LiCl (A). Green stained actin appears along the hyphae, especially in the hyphal tips. Red stained Spitzenkörper (Spk) appears in the hyphal tip using Synapto-red FM 4-64. On the other hand, less actin dynamics in *S. commune* using Life-act GFP with LiCl treatment (B). Green stained actin appears along the hyphae, but not in hyphal tips. No red stained Spitzenkörper is visible in the hyphal tip using Synapto-red FM 4-64.

Here, time lapse studies in control hyphae and lithium treated *S. commune* 15-2A using Life-act GFP were performed in different time intervals (Figure 20).

Low dynamic of actin was observed in the fungus with LiCl treatment. Meanwhile, in the control hyphae, a highly dynamic movement of actin was observed. Also, a relatively high abundance of actin was found along the hyphae and a dense accumulation of actin in hyphal tips. Further, Spitzenkörper could only be observed in control hyphae as dark apical body using SynaptoRed FM 4-64, whereas in LiCl treatment this prominent structure was not visible. In the wild-type, hyphal growth is polarized and managed by Spitzenkörper which is surrounded by actin.

Together with many vesicles translocated in the tips, actin moved dynamically and at some point, actin patches dispersed. This specific phenomenon was not found in the lithium treated fungus. The actin patches in lithium treated fungus did not actively move as it was shown in the control. In control, instead of located in the apical hyphae, actin patches were rather distributed well along the hyphae. In some cases, hyphae from the lithium treatment did not appear straight as it is shown in the control.

### 3.6 Inositol monophosphatase and inositol monophosphatase-like gene family

Inositol monophosphatase gene *imp* comprises of 1247 bp, which encodes 340 amino acid and it is localized at scaffold 2:3726779-3728025 in the third version of *S. commune* genome JGI schco3. It has been found that there are nine putative proteins which are similar with the protein that encodes inositol monophosphatase gene (*imp*). Despite of relatively high similarity compared to the *imp*, these nine candidates, however does not have high similarity in the three unique motifs named motif A, motif B, and motif C.

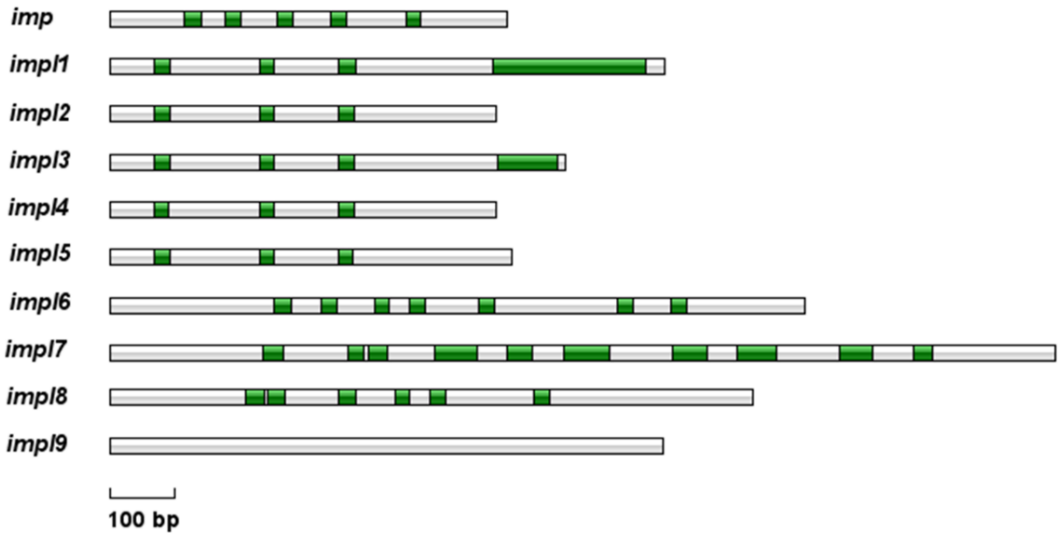


Figure 21. Exon and Intron distribution of *imp* and *imp* like gene family visualized using Illustrator for Biological Sequences (IBS). White boxes indicate exons and green boxes indicate introns. Protein IDs of the genes are listed as following: *imp* (schco3 protein ID 2697390), *impl1* (schco3 protein ID 2506241), *impl2* (schco3 protein ID 2522254), *impl3* (schco3 protein ID 2603869), *impl4* (schco3 protein ID 2669086), *impl5* (schco3 protein ID 2702509), *impl6* (schco3 protein ID 2567698), *impl7* (schco3 protein ID 2615498), *impl8* (schco3 protein ID 2635183), *impl9* (schco3 protein ID 2690717).

These three motifs are important for the enzyme activity. For this reason, the nine candidates are named as inositol monophosphatase-like gene.  High similarity has been found among *impl1, impl2, impl3, impl4*, and *impl5*. The introns and exons distribution among these five genes are highly identic, particularly in *impl2*, *impl4* and *impl5* (Figure 21). These five inositol monophosphatase like genes possess a salt-sensitive 3'-phosphoadenosine-5'-phosphatase HAL2/SAL1 domain. On the other hand, *impl6, impl7, impl8* and *impl9* encompass histidine acid phosphatase (HAP) domain. HAP domain is typical for inositol polyphosphate phosphatase and known to be responsible for inositol hexakisphosphate dephosphorylation into lower species of inositol phosphates.

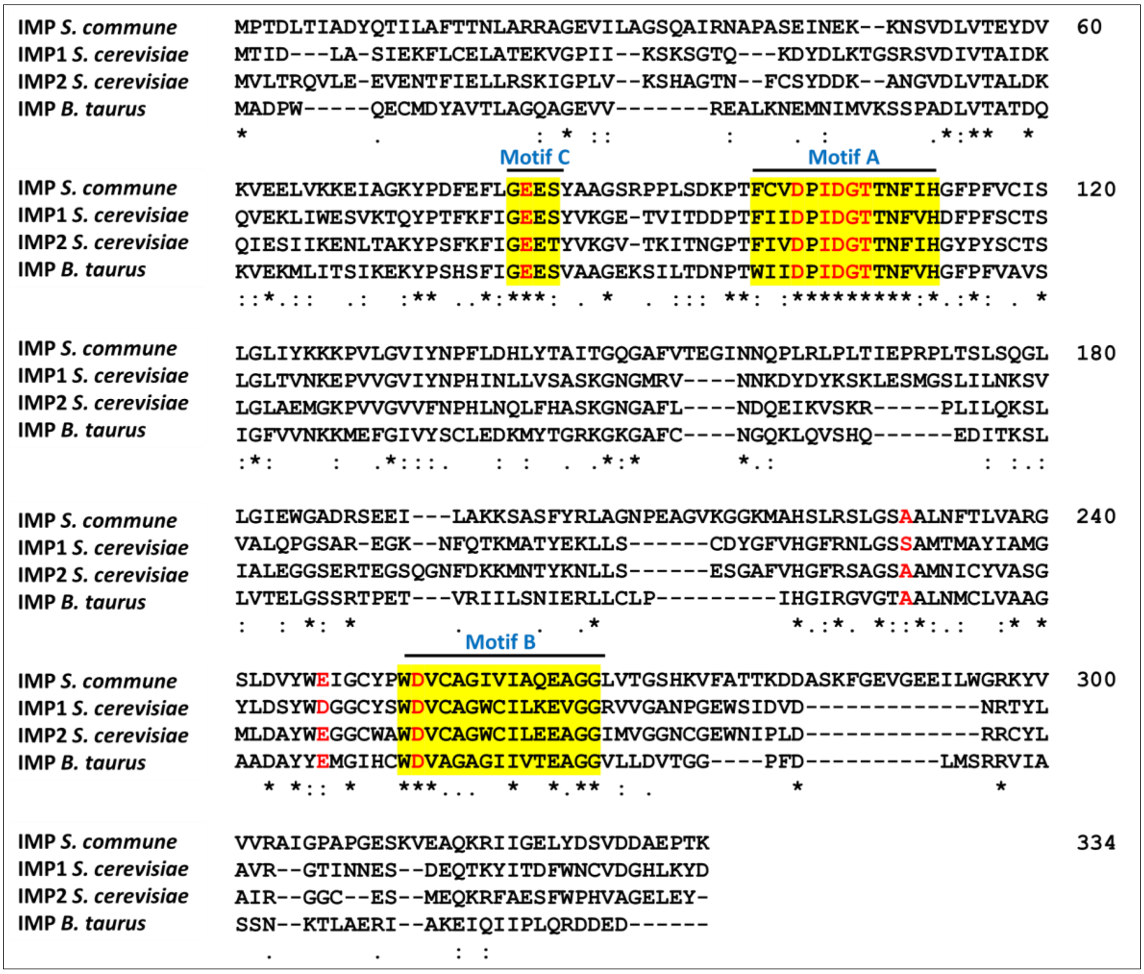
## 3.7 Inositol monophosphatase

*S. commune* inositol monophosphatase (Schimp) protein structure was compared to bovine inositol monophosphatase (Btimp), which the three-dimensional structure was previously examined and identified. These three unique motifs are known as a lithium binding sequence. Important to note that Btimp sequence was well identified, the crystal structure was determined and functions of individual amino acids was confirmed by site-directed mutagenesis. The three motifs are known to be the key active site interactions, that comprises of identified amino acids involved in the putative metal binding and nucleophilic activation interaction (Table 12).

The common substrate for inositol monophosphatase are  inositol 1-monophosphate, inositol 3-monophosphate and inositol 4-monophosphate, thus the prediction that there are three hydroxyl groups that makes four hydrogen bonds interaction with the enzyme, namely, 2-hydroxyl (2-OH) , 4-hydroxyl (4-OH) and 6-hydroxyl (6-OH). Inositol 2-OH binds to $D^{93}$ and $A^{196}$, 4-OH binds to $E^{213}$,  and 6-OH binds to $D^{220}$. The phosphate group was predicted to bind the $G^{94}$ and $T^{95}$ and in addition, the phosphate group is positioned at alpha-helical of residues 95-100 located in the C-terminal. The metal binding site 1 is necessary for the following phosphate binding substarte by activating $H_2O$ that is crucial for nucleophilic attack on the phosphate phosphorous. On the other hand, the metal binding site 2 was predicted to be a site for phosphate binding following a substrate hydrolysis that denotes the putative lithium inhibition. Metal binding 3 is also necessary for the following phosphate binding substarte, however its binding is considerably weak and not cooperative.

**Table 12. Key active sites interaction for substrate, metal binding, and mechanistic interaction of *Schizophyllum commune* inositol monophosphatase**

| Amino acid residue | Binding capability |
|---|---|
| $E^{70}$ | site 1 metal, nucleophilic $H_2O$, and site 3 metal |
| $D^{90}$ | site 1 metal, site 2 metal |
| $I^{92}$ | site 1 metal |
| $D^{93}$ | site 2 metal and substrate 2-OH |
| $G^{94}$ | substrate phosphate |
| $T^{95}$ | substrate phosphate and possible nucleophilic $H_2O$ |
| $A^{196}$ | substrate 2-OH |
| $E^{213}$ | substrate 4-OH |
| $D^{220}$ | site 2 metal, substrate 6-OH |



```
IMP S. commune       MPTDLTIADYQTILAFTTNLARRAGEVILAGSQAIRNAPASEINEK--KNSVDLVTEYDV   60
IMP1 S. cerevisiae   MTID---LA-SIEKFLCELATEKVGPII--KSKSGTQ---KDYDLKTGSRSVDIVTAIDK
IMP2 S. cerevisiae   MVLTRQVLE-EVENTFIELLRSKIGPLV--KSHAGTN--FCSYDDK--ANGVDLVTALDK
IMP B. taurus        MADPW-----QECMDYAVTLAGQAGEVV-------REALKNEMNIMVKSSPADLVTATDQ
                     *         .           : * ::        :    . :       .*:**  *
                                           Motif C                Motif A
IMP S. commune       KVEELVKKEIAGKYPDFEFLGEESYAAGSRPPLSDKPTFCVDPIDGTTNFIHGFPFVCIS   120
IMP1 S. cerevisiae   QVEKLIWESVKTQYPTFKFIGEESYVKGE-TVITDDPTFIIDPIDGTTNFVHDFPFSCTS
IMP2 S. cerevisiae   QIESIIKENLTAKYPSFKFIGEETYVKGV-TKITNGPTFIVDPIDGTTNFIHGYPYSCTS
IMP B. taurus        KVEKMLITSIKEKYPSHSFIGEESVAAGEKSILTDNPTWIIDPIDGTTNFVHGFPFVAVS
                     ::*.::  .:  :** ..*:***: . *  . ::: **: :*********:*.:*: . *

IMP S. commune       LGLIYKKKPVLGVIYNPFLDHLYTAITGQGAFVTEGINNQPLRLPLTIEPRPLTSLSQGL   180
IMP1 S. cerevisiae   LGLTVNKEPVVGVIYNPHINLLVSASKGNGMRV----NNKDYDYKSKLESMGSLIILNKSV
IMP2 S. cerevisiae   LGLAEMGKPVVGVVFNPHLNQLFHASKGNGAFL----NDQEIKVSKR-----PLILQKSL
IMP B. taurus        IGFVVNKKMEFGIVYSCLEDKMYTGRKGKGAFC----NGQKLQVSHQ------EDITKSL
                     :*:    :  .*:::.   : :  . .*:*       *.:               : :.:

IMP S. commune       LGIEWGADRSEEI---LAKKSASFYRLAGNPEAGVKGGKMAHSLRSLGSAALNFTLVARG   240
IMP1 S. cerevisiae   VALQPGSAR-EGK--NFQTKMATYEKLLS------CDYGFVHGFRNLGSSAMTMAYIAMG
IMP2 S. cerevisiae   IALEGGSERTEGSQGNFDKKMNTYKNLLS------ESGAFVHGFRSAGSAAMNICYVASG
IMP B. taurus        LVTELGSSRTPET---VRIILSNIERLLCLP---------IHGIRGVGTAALNMCLVAAG
                     :  : *: *       .     . .*              *.:*. *::*:.:  :* *
                                 Motif B
IMP S. commune       SLDVYWEIGCYPWDVCAGIVIAQEAGGLVTGSHKVFATTKDDASKFGEVGEEILWGRKYV   300
IMP1 S. cerevisiae   YLDSYWDGGCYSWDVCAGWCILKEVGGRVVGANPGEWSIDVD-------------NRTYL
IMP2 S. cerevisiae   MLDAYWEGGCWAWDVCAGWCILEEAGGIMVGGNCGEWNIPLD-------------RRCYL
IMP B. taurus        AADAYYEMGIHCWDVAGAGIIVTEAGGVLLDVTGG----PFD----------LMSRRVIA
                       * *:: *   ***..  *  *.** : .          *              *

IMP S. commune       VVRAIGPAPGESKVEAQKRIIGELYDSVDDAEPTK                          334
IMP1 S. cerevisiae   AVR--GTINNES--DEQTKYITDFWNCVDGHLKYD
IMP2 S. cerevisiae   AIR--GGC--ES--MEQKRFAESFWPHVAGELEY-
IMP B. taurus        SSN--KTLAERI--AKEIQIIPLQRDDED------
                       .       .     : :
```

Figure 22. Multiple sequence alignment of *S. commune*, *S. cerevisiae*, and bovine (*Bos taurus*) inositol monophosphatase amino acid sequences. The key substrate, metal binding, and interaction site based on the bovine enzyme crystal structure are red-marked and explained in more detail in Table 12. The crucial regions highlighted in yellow shows the motif A, B and C that are known to be important for the substrate and metal binding, as well as nucleophile activation. At least one of these motifs can be found in following enzymes inositol polyphosphate 1-phosphatase and fructose 1,6-bisphosphatase. Inositol polyphosphate 1-phosphatase is an enzyme that is responsible to dephosphorylate inositol 1,3,4-trisphosphate and inositol 1,4-bisphosphate.

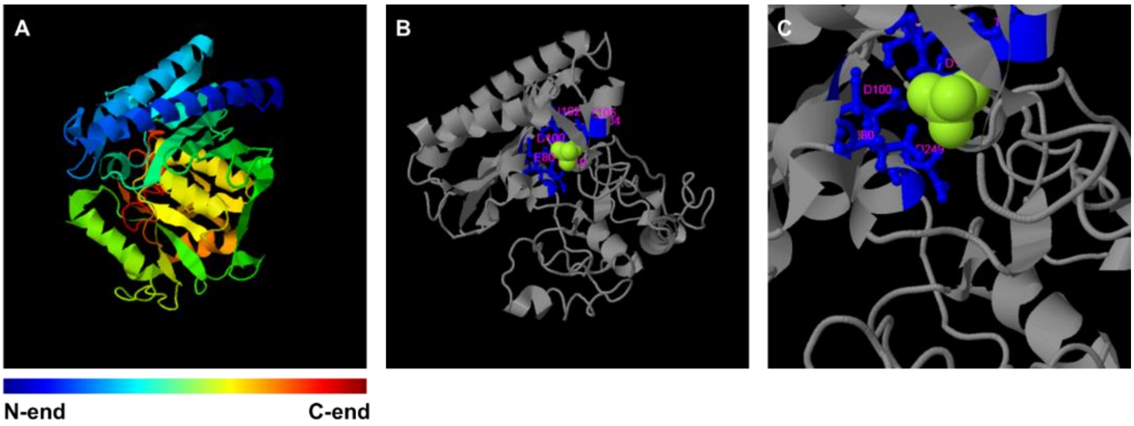


Figure 23. 3D models of inositol monophosphatase was generated using the I-TASSER server. Inositol monophosphatase protein model (colored from N- to C-end) is similar to 3D structure from IMP of *Bos taurus* (A). Predicted ligand binding site (B). The magnified version of predicted ligand binding site (C). The greens sphere shows the predicted binding ligand and blue is the binding residues.

### 3.8 Comparative genetic map of *imp* and surrounding genes

The analysis of chromosomal organization of *imp* and surrounding genes was done using the published genome data from three different *S. commune* strains. Strain H4-8, the first strain which its genome was published, was used as reference for the analysis. The genome of strains Loenen D/ LoeD and Tattone D/ TatD were published several years later. The two latter strains were included in the analysis to investigate whether the inositol monophosphatase gene (*imp*) and surrounding genes are highly conserved in every strain. The inositol monophosphatase gene is located in scaffolds 1 in the case of strain H4-8, in scaffold 30 in strain LoeD, and in scaffold 181 in strain TatD. The analysis and visualization of the genetic map were performed manually using ChromoMapper. The predicted gene, size, and function data were obtained from JGI genome. In the reference strain H4-8, *imp* is localized in scaffold 1: 3366114-3367360 surrounded by Conserved Zinc finger (*znf*) and molecular chaperone DNAJ superfamily (*dnaj*). Following genes including *fabG* which encoding glucose/ribitol dehydrogenase, *caf1* encoding nucleosome remodeling, *psma1* encodes for 20s proteasome, and *shl* encodes for chromatin remodeling protein, are in close proximity with *imp*.

Conservation between strains H4-8 and LoeD was high in most cases, but less similar compared to TatD. The two strains LoeD and TatD possess similar gene location and similar transcriptional direction in comparison to the strain H4-8. Important to notice that the scaffolds where *imp* is located, has different length in every strain, TatD has the shortest scaffolds amongst the strains. The reason for this could be different sequencing methods used for the genome analysis. Conserved Zinc finger (*znf*) and molecular chaperone DNAJ superfamily (*dnaj*) appeared to be the neighboring gene of *imp*. This arrangement was also found in human *imp*. Underlining the similarity between the H4-8 strains and LoeD chromosomal organization, thirteen genes were found to have high identity and arranged in the same manner (*ipo9, bt, caf1, znf, imp, dnaj, psma1, shl, fip1, rabgap1, and rnh*) and only three genes (*cog*, *pnc1* and *sdr*) have different orders however the rest are localized in the same manner.

In TatD strain is also similar to other strains. Nine genes possess similar chromosomal localization (ca*f1, znf, imp, dnaj, psma1, shl, fip1, rabgap1,* and *rnh*) and two other genes have different orders in the chromosomal mapping (*pnc1* and *sdr*). Interestingly, the high identity of *imp* was found in both strains, 97,6 % and 98,2 % LoeD and TatD, respectively.

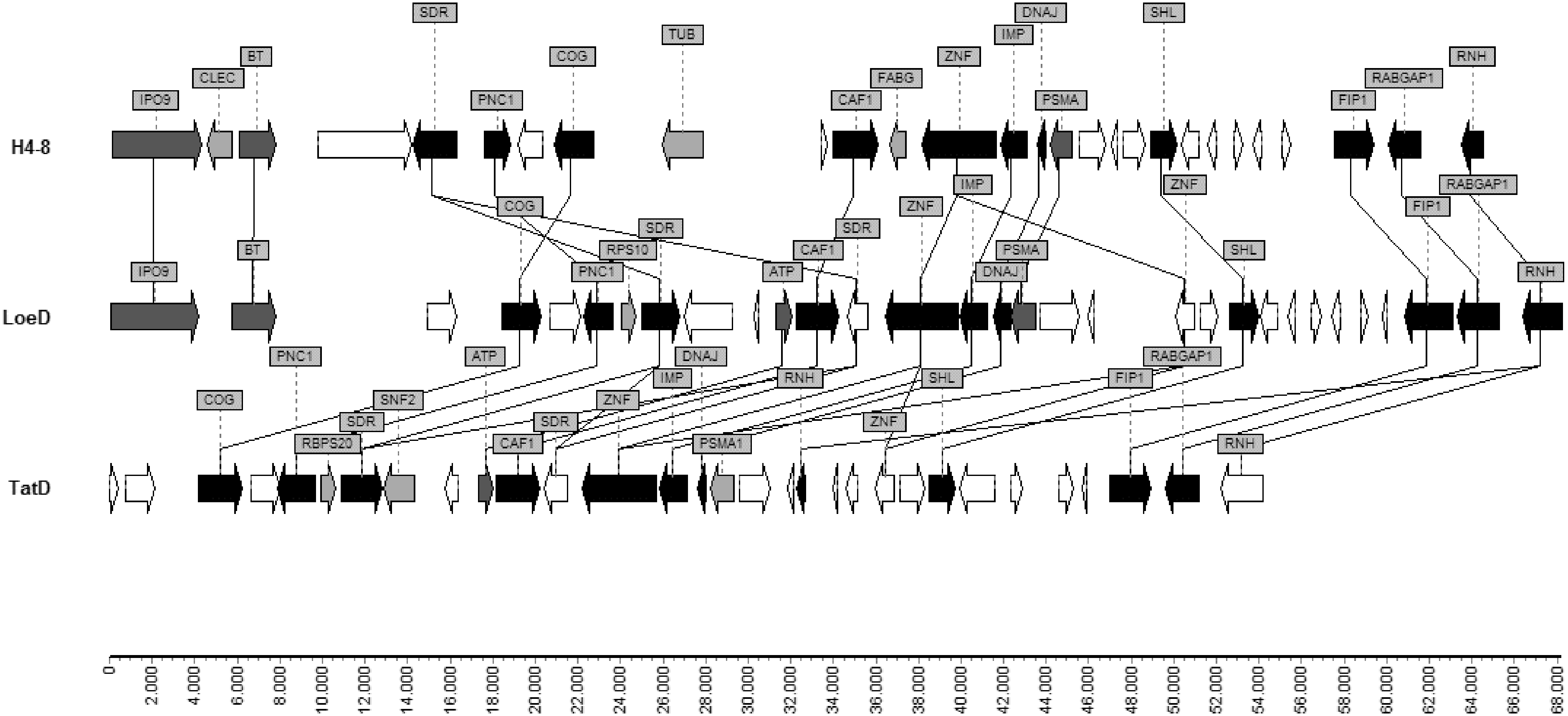


Figure 24. Schematic comparison of *imp* surrounding genes in three different strains H4-8, LoeD, and TatD. H-48 was used as reference strain. The strain names are labelled on the left side of the figure. Inositol monophosphatase gene and surrounding genes are depicted in this figure. The *imp* and surrounding genes were localized in different scaffolds, scaffold 1 for the strain H4-8, scaffold 30 for the strain Loenen D/LoeD and scaffold 181 for the strain Tattone D/ TatD. The white arrows indicate the unknown genes. Black arrows show that the corresponding genes are found in three genomes.

## 3.9 Overexpression of inositol monophosphatase (*imp*)

### 3.9.1 Validation of *imp* overexpression

Inositol monophosphatase (*imp*) gene was overexpressed under *tef* promoter. This was done by fusing the *tef* promoter with the target gene *imp*. Both *tef* promoter and *imp* sequence were generated from PCR and the fusion of these two products was facilitated via yeast recombination. The fusion product was excised and cloned into a plasmid containing the selection marker tryptophan, called pTrp, which later resulted in a new plasmid, pOEIMP.

*S. commune* T33 was transformed with plasmid pOEIMP. *S. commune* T33 is a tryptophan auxotroph strain, thus it was selected as parental strain that is suitable for tryptophan selection. Two *imp* overexpressing mutants were obtained and thus named OEIMP4 and OEIMP6 and in addition a control for transformation was generated by transforming *S. commune* T33 with an empty vector with no fusion product of *tef* promoter and *imp*, thus it is labelled as empty vector control (EVC1). To proof if the transformation was successful, PCR using a primer pair that span the fusion of *tef* promoter and *imp* was carried out. The expected amplicon is 1.2 kb, was proven and it is shown in figure 25. The positive bands were cut and sent for sequencing analysis.

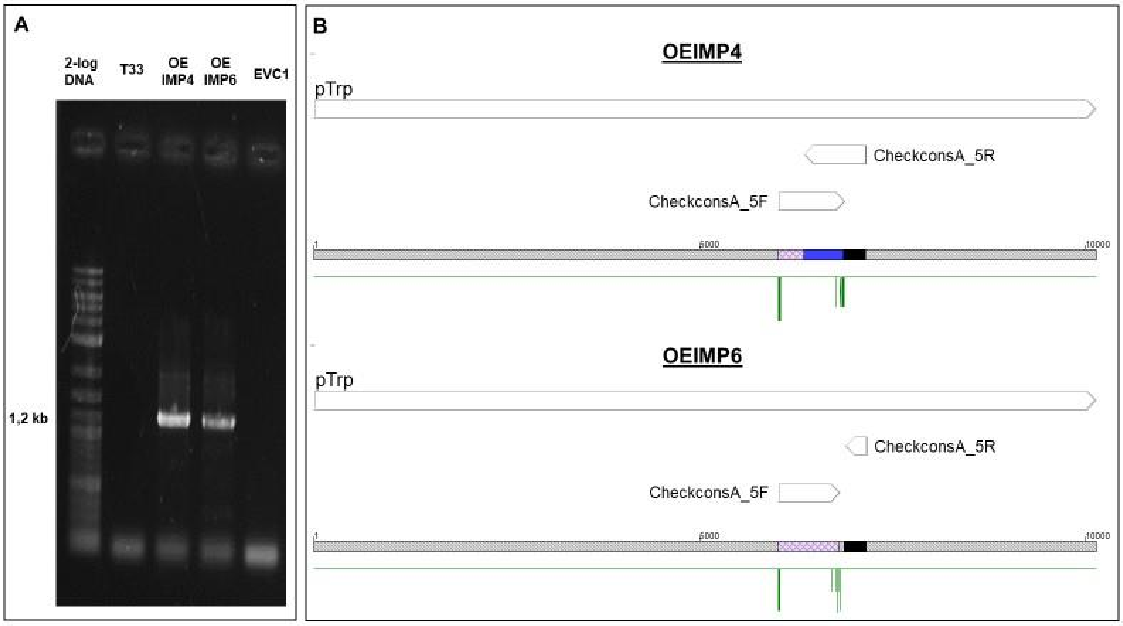


Figure 25. Verification of *imp* overexpression fragment by PCR in two *imp* overexpressing mutants, OEIMP4 and OEIMP6 using a primer pair to check the fusion of *tef* promoter and *imp* in pTrp (CheckconsA F and CheckconsA R). Electrophoresis gel picture of fusion *tef* promoter and *imp* fragment. Molecular weight marker used is 2-log DNA; T33 is parental strain; EVC1 is empty vector control and *imp* overexpressing mutants are OEIMP4 and OEIMP6 (A). Contig of TEF promoter and IMP fusion product of OEIMP4 and OEIMP6. The PCR amplicon was sequenced, and the sequences separately were aligned with the plasmid pOEIMP sequence (B).

Figure 25 A shows amplicon from OEIMP4 and OEIMP6 showing the expected size and the control strains T33 and EVC1 showing no amplicon. Thus, indicating that OEIMP4 and OEIMP6 are successful transformants. Furthermore, the expected alignment was compared to the pOEIMP sequence. The contig in Figure 25 B shows the right alignment exactly in the fusion of *tef* promoter and *imp* area. The gene expression of *imp* of these two putative transformants, OEIMP4 and OEIMP6 were further analyzed using RT-qPCR to determine the higher expression of *imp* compared to control strains. Gene expression of *imp* was normalized to the expression of housekeeping genes *tef*, *ubi* and *act*.

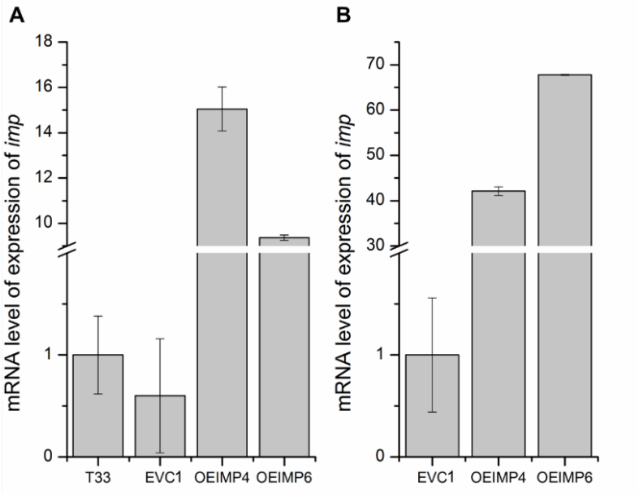


Figure 26. mRNA level expression of *imp* overexpressing mutants OEIMP4 and OEIMP6 as well as the control strain (T33 - parental strain and EVC1- empty vector control). A. T33 as the control strain and B. EVC1 as the control strain. In both graphs are clearly seen that the imp gene transcriptomic level is higher expressed in comparison to both control strains.

The expression of *imp* gene from the *imp* overexpressing mutants were compared separately with either T33 as parental strain (Figure 26 A) or to EVC1 as empty vector control strain (Figure 26 B). Both comparisons show that *imp* transcriptomic level expression is elevated in both *imp* overexpressing mutants. To compare the expression with control strain, it is important to notice that control strain always has a ratio 1. Ratio starting from 2 folds or -2 folds shown significant difference to the control strain. The *imp* gene of OEIMP4 and OEIMP6 strains in comparison with T33 were expressed higher within 15-folds and 9-folds, respectively. There is a remarkable increase of *imp* gene expression of OEIMP4 and OEIMP6 in comparison to the EVC1. High expression of *imp* gene up to 42-folds and 67-folds in OEIMP4 and OEIMP6, respectively, was observed in the *imp* gene expression level compared to the EVC1.

### 3.9.2 Characterization of *imp* overexpressing mutants

The growth of *imp* overexpressing mutants was determined and compared to parental strain and to empty vector control strain. Cultivation and analysis were performed for 9 days in two different media, CYM-T and MM-UT (Figure 27). Supplementation of tryptophan is required for the parental strain since T33 (parental strain) is tryptophan auxotroph, thus the supplementation of tryptophan is required also for other strains. The fungal growth of all strains on MM-UT is relatively low in comparison to the cultivation of all strains in CYM-T. There was no significant difference in growth rate between the fungal strains. Therefore, we can conclude that *imp* overexpression does not affect fungal growth significantly. There was also no difference in morphology between all strains. In MM-UT, the aerial mycelial growth was less developed in all fungal strain. Some hyphal sectors grew faster than others, resulting in an irregular mycelial colony shape which was shown in all fungal strains cultivated in MM-UT.

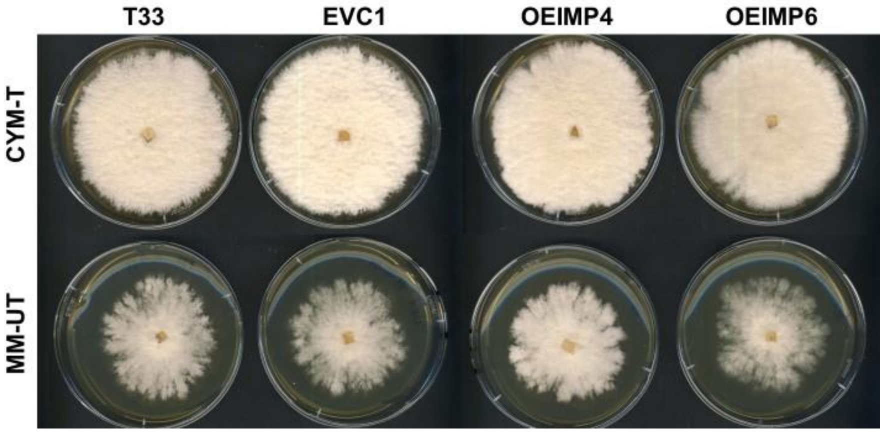


Figure 27. Morphology of *imp* overexpressing mutants OEIMP4 and OEIMP6 along with the control strain (T33 - parental strain and EVC1- empty vector control) in two different media, CYM-T and MM-UT. The age of cultivation shown in this picture is 10 days.

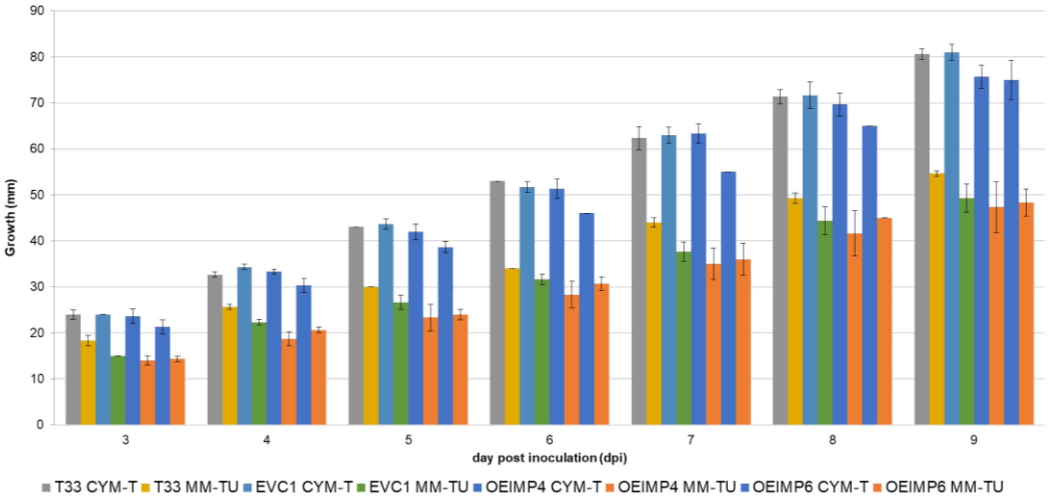


Figure 28. Growth of overexpressing *imp* mutants and the parental strain as well as the empty vector control strain. There is no significant difference of growth rate between all strains, either in CYM-T or MM-TU.

Occasionally multi-branching was found in all fungal strains cultivated in MM-UT under bright-field microscopy investigation. These suggest that the overexpressing mutant has no unique characteristics in terms of growth. The growth rate of T33, EVC1, OEIMP4 and OEIMP6 in CYM-T are 89 mm/day, 90 mm/day, 85 mm/day and 84 mm/day, respectively. Slower growth rate of *imp* overexpressing mutants was shown in the nutrient limited media MM-UT. The average growth reduction in MM-UT fungal cultivation is up to forty percent. The growth rate in MM-UT of T33, EVC1, OEIMP4 and OEIMP6 are 56 mm/day, 55 mm/day, 55 mm/day, and 56 mm/, respectively (Figure 28).

### 3.9.3 Mating interaction of *imp* overexpressing mutants

Mating interaction involving *imp* overexpressing strains was performed with some selected compatible strains (1-106, 1-69, T-50, 4-40, 12-43, and T-42, representative pictures are only the ones from the mating interaction with 1-106, 1-69, T-50, see Figure 29). This experiment was performed in order to investigate the effect of *imp* overexpression on mating in *S. commune.* The mating behavior of *the imp* overexpressing strains was observed until mature fruiting bodies were formed. Clamp formation in all mating plates could be observed after the mating interaction of the two compatible mating strains. None of the mating interactions show a peculiar phenomenon. However occasionally three clamps cell in a close proximity were observed in some mating strains interacting with *imp* overexpression. This phenomenon is very rare, so it cannot be concluded, that this occurrence is solely because of the *imp* overexpression.

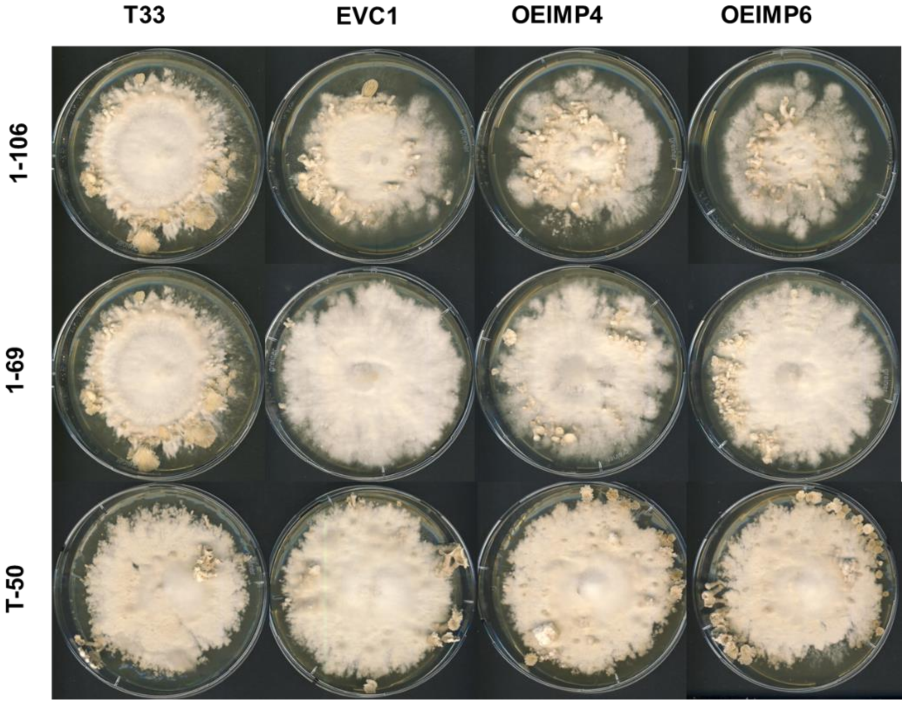


Figure 29. Mating interaction of *imp* overexpressing mutants OEIMP4 and OEIMP6 as well as control strain (T33 - parental strain and EVC1- empty vector control) with the three different compatible mating strains (1-106, 1-69 and T-50). The age of fungal cultivation in this picture is 17 days.

The overexpression mutant of *imp* crossed with 1-106 and 1-69 developed and produced more fruiting bodies in comparison to the EVC1 strains, however the fruiting bodies from the *imp* overexpressing mutants mostly did not develop well as it is visible in the parental strains' fruiting bodies. Several fruiting bodies produced from the mating interaction with the parental strain T33 show some well-developed lamellae ornamented with the typical split-gills facing towards the lid of petri dish. Tremendous amount of fruiting body production was also observed in the mating interaction between *imp* overexpression and the strain T-50. The difference with the other two compatible strains mentioned above is that some well-developed lamellae could be observed in the produced fruiting bodies. From previous study, there was indication that inositol may stimulate the production of fruiting bodies (Xie *et al.*, 2017). If there is a tremendous amount of fruiting bodies, the nutrition for each fruiting bodies on the plate is limited. This is the reason why the fruiting bodies were not fully developed in the plates that has a greater amount of fruiting bodies, since there might be a competition in nutrients uptake from the media.

### 3.9.4 Inositol phosphates profile in *imp* overexpressing mutants

Inositol phosphates analysis was performed in inositol monophosphatase overexpressing mutants and compared to the empty vector control (EVC1) strain. From the HPLC profile (Figure 30), the entire inositol phosphates were significantly increased, except the dephosphorylated products of $IP_6$, which are $IP_{5b}$ and $IP_{5c}$.

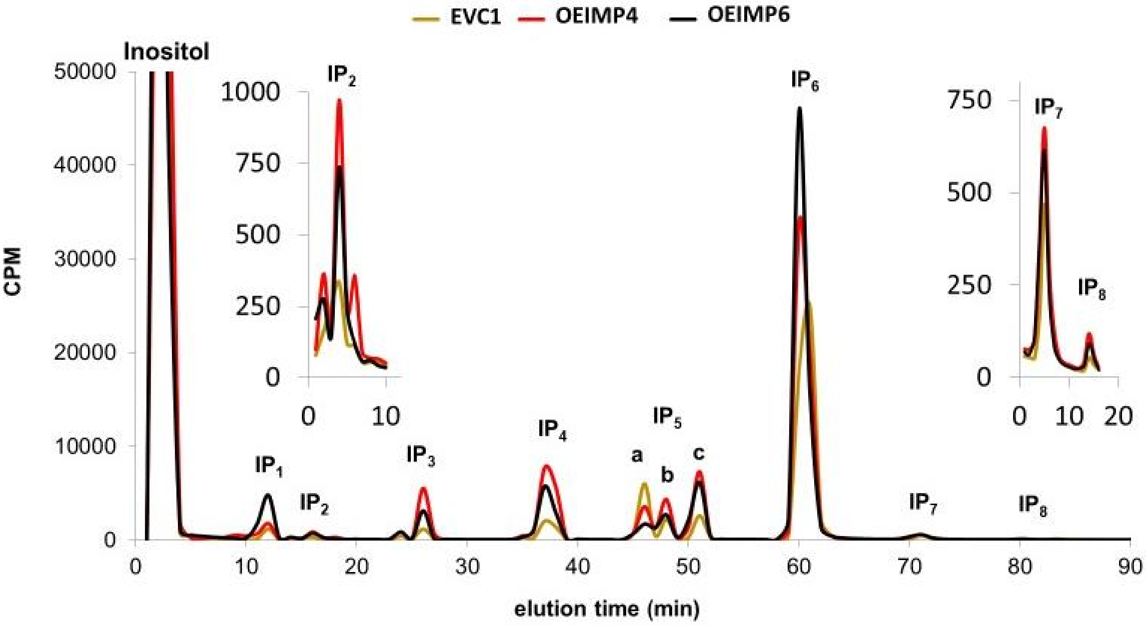


Figure 30. Strong Anion Exchange-High Liquid Performance Chromatography (SAX-HPLC) elution profile of soluble inositol phosphates of *S. commune* overexpressing mutants and empty vector control (EVC1). Mycelia were labeled with myo-[2-$^3$H(N)]-Inositol for 24 h before total inositol phosphate extraction and SAX-HPLC analysis. The chromatogram of inositol bisphosphate ($IP_2$) and inositol pyrophosphate $IP_7$ -$IP_8$ were magnified in a different scale.

Inositol is expected to be elevated since the inositol monophosphatase was overexpressed. However, from the ratio calculation compared to the inositol phosphates belong to the EVC1, it is shown that inositol is not elevated (Figure 31). The reason for this might be because of difficulty in washing step during inositol phosphate extraction. Liquid MM-U could not be washed completely, presumably leaving the trace of $^3$H-Inositol in the extract. This presumption is supported with the fact that the inositol phosphates are elevated in the *imp* overexpressing strains. In the HPLC profile, it is shown that $IP_{5a}$, $IP_{5b}$, $IP_6$, and $IP_7$ were reduced in *imp* overexpressing mutants. $IP_{5b}$ and $IP_{5c}$ are the dephosphorylation product from the full phosphorylated inositol phosphate $IP_6$. $IP_{5a}$ is on the other hand, a precursor of for $IP_6$ production.

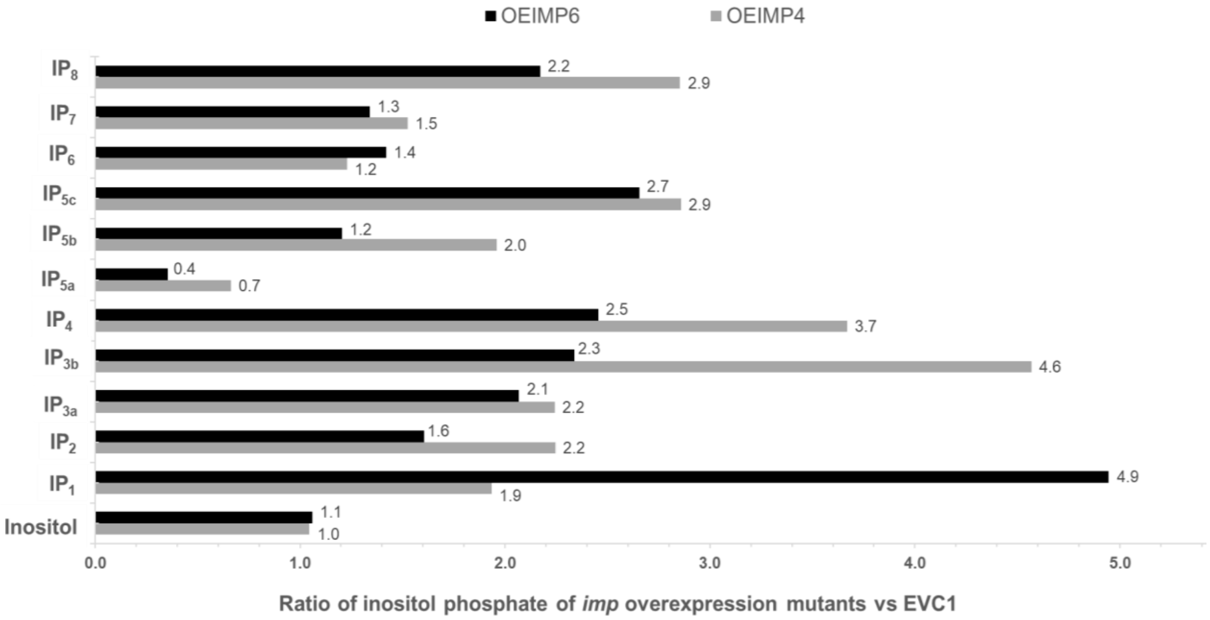


Figure 31. Ratio of inositol phosphate of *imp* overexpressing strains vs EVC1. The entire inositol phosphate species were elevated in *imp* overexpressing mutants OEIMP4 and OEIMP6, except $IP_{5a}$, $IP_{5b}$, and IP6 and.

The highest elevation in OEIMP4 are $IP_{3b}$ and $IP_4$, and the highest elevation for the OEIMP6 was $IP_1$ (Figure 31). The increment of low inositol phosphate species in *imp* overexpressing strains indicates that the building block of inositol phosphate, which is inositol, might be as well increased, however the increase of inositol was not shown in HPLC analysis. Inositol monophosphatase is the key enzyme for the preservation of the inositol production. It controls the cellular availability of inositol, affecting inositol phosphate metabolism and phosphatidyl inositol signaling. As it was argued above, there might be an obstacle in washing, showing a false result in inositol level measurement.

### 3.9.5 Effect of SDS on *imp* overexpressing mutants

Inositol is a basic molecule that is used to build a phosphatidylinositol which plays a crucial role in sustaining the cell membrane integrity. The effect of sodium dodecyl sulfate (SDS) was tested on the *imp* overexpressing mutants, parental strain and empty vector strain. The *imp* overexpressing mutants were less susceptible to 150 µg/ml SDS in comparison to the control strains. Higher concentration of SDS (200 µg/ml) was also tested and the result shows that the higher concentrated of SDS, the lesser the growth of the fungi (Figure 32). In T33 and EVC1, some abnormal hyphal growth was observed under microscope. The hyphae grew wavy instead of straight, but however this phenomenon appear only in some mycelial section, where the aerial mycelia have less growth compare to other sections. This typical feature could not be observed in *imp* overexpressing mutants OEIMP4 and OEIMP6. This evidence leads to the speculation that *imp* overexpressing mutants possess stronger cell membrane due to the formation of more phosphatidylinositol in the cell membrane. Other cell membrane stressors or antifungal such as Tebuconazole and Fenchlorazole were tested against *imp* overexpressing mutants and control strains. However, there is no significant difference in growth rate among all strains in the presence of Tebuconazole and Fenchlorazole (Traxler, 2017).

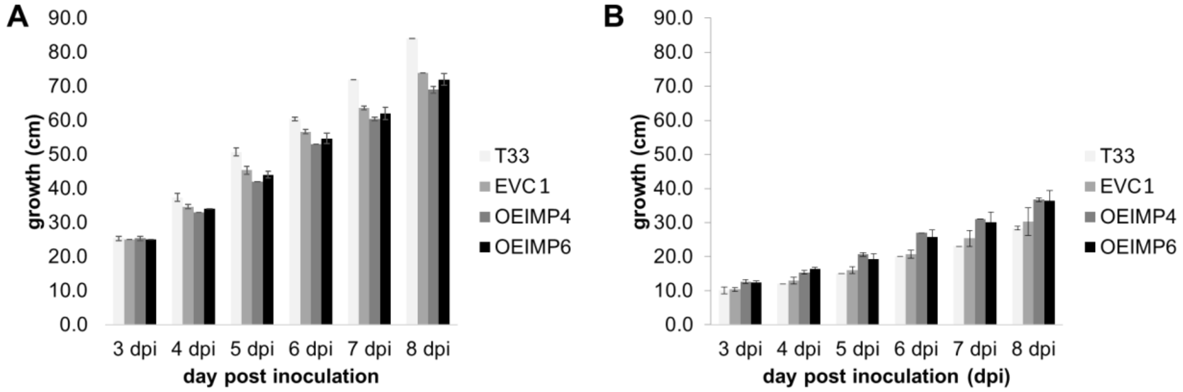


Figure 32. Growth of T33, EVC1, OEIMP4 and OEIMP6 without SDS addition (A) and growth of T33, EVC1, OEIMP4 and OEIMP6 with 150 SDS presence µg/ml (B). Overexpression of *imp* mutants are less susceptible to SDS in comparison to the parental strain and EVC1 strain.

### 3.10 Proteome analysis in *imp* overexpressing mutants

LC-MS based proteome analysis of *imp* overexpressing mutants shows the induction of 43 proteins that correspond to cellular processes and signaling, 41 proteins that are responsible for information storage and processing, 58 proteins are involved in the metabolism, 31 poorly characterized proteins, and 114 unidentified proteins based on EuKaryotic Orthologous Groups (KOG) group. In total there are 287 proteins that are induced in response to *imp* overexpression (Figure 33). One of the most striking features is the induction of membrane trafficking machinery in response to *imp* overexpression, which will be explained in another section (see 3.11). Furthermore, *imp* overexpression might also stimulate another G-protein related signaling pathway. Ras GTPase was decreased within 2.79-folds. As it is hypothesized that there is an unknown mechanism crosstalk between Ras and inositol signaling. The cyclic AMP signaling machinery such as two putative guanine nucleotide binding protein (G-protein) alpha subunit and adenylate cyclase-associated protein (CAP) were induced within 2.37-folds, 3.71-folds, and 3.01-folds. CAP is involved in the organization of actin microfilament that is located in the plasma membrane. Moreover, CAP is necessary for the activation of cAMP signaling through adenylate cyclase activation in fruiting bodies formation, vesicle trafficking and endocytosis. As it is expected the inositol monophosphatase is also strongly induced at 26.98-folds, indicating that the overexpression of *imp* was successfully performed, and the up-regulation occurs not only on the transcriptome level but also on the proteome level.

Another protein called septin which is important for controlling the filamentous morphology of fungi is also found up-regulated. Septin is a GTP binding protein which is required for actin formation and chitin-ring formation. Therefore, it is also not surprising to observe the induction of chitinase in the proteome level, since septin stimulates the formation of chitin and the accumulation of chitin will eventually result into production of chitinase to aid in chitin degradation. It is important also to mention that a protein possessed histidine acid phosphatase (HAP) was induced within 3.06-folds. As we have found in the earlier study, that the attenuation of inositol phosphate by lithium affect the repression of HAP. The counter effect on *imp* overexpression, which is the induction of protein possessed HAP domain, was observed.

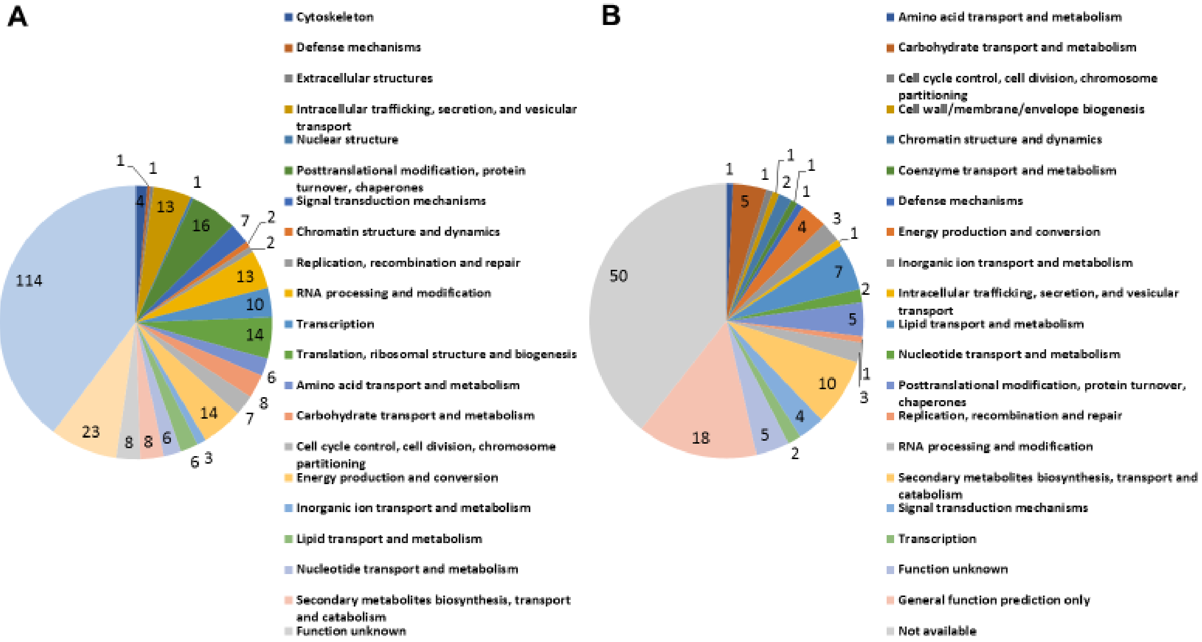


Figure 33. Classification of protein regulated in OEIMP4 strain versus empty vector control (EVC1) strain based on EuKaryotic Orthologous Groups (KOG) class. There are 287 induced proteins (A) and 127 repressed proteins (B) prior to *imp* overexpression.

HAP is a domain possessed by inositol polyphosphate phosphatases, such as inositol hexakisphosphate phosphatase that is responsible to convert $IP_6$ to $IP_5$. This evidence is convincing, since inositol the building block of inositol phosphates is presumed to be elevated as an effect of *imp* overexpression. This means the higher inositol level in the cell; the more inositol phosphates could be generated and therefore inositol phosphate phosphatases activation is stimulated. On the other hand, there were 127 repressed proteins prior to *imp* overexpression. The dominant group of proteins which were repressed in *imp* overexpressing mutant OEIMP4, were involved the secondary metabolites biosynthesis, transport and catabolism such as flavin-containing monooxygenase, cytochrome P450, phenylalanine and histidine ammonia-lyase, ATPase, and lipocalin. To check whether the protein kinase C (PKC) is regulated prior to *imp* overexpression, we did analysis in PKC. Unfortunately, there is no significant difference of PKC between *imp* overexpressing mutants and the control strains (Figure S5).

### 3.11 Membrane trafficking related proteins are induced in *imp* overexpressing strains

Proteome analysis reveals the induction of 18 proteins associated with membrane trafficking. Four proteins are associated with the cytoskeleton, three of them are actin binding protein-related proteins and another one is kinesin-like protein, were found induced in overexpressing mutant OEIMP4. Actin-based cytoskeleton plays a critical role in fungal growth and development by maintaining cell shape, structural support and cell polarity which are critical for maintenance of the polar growth, cell cycle progression, mitosis, and meiosis. The actin-based cytoskeleton proteins support the endocytosis and exocytosis process. Actin dynamics facilitate membrane fusion and merge fusing vesicles. The other fourteen proteins are the proteins which are responsible for intracellular trafficking, secretion and vesicular transport, including signal recognition particle SRP19, ABC transporter, C5 cytosine-specific DNA methylase, acyltransferase, signal recognition particle SRP72, mitochondrial import inner membrane translocase TIM8 containing zinc finger domain, armadillo type-fold SEC7-like, VPS5/SNX1 containing Phox domain, cysteine protease required for autophagy Apg4p/Aut2p, nuclear transport factor 2, t-SNARE, dynamin/VPS1, and two SNF7 proteins.

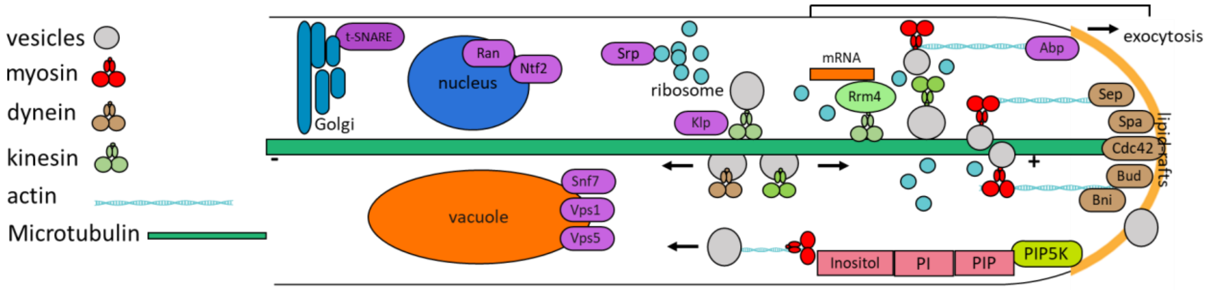


Figure 34. Membrane trafficking machinery modified after Steinberg (2007). The overexpression of inositol monophosphatase stimulates the activation of several proteins related to membrane trafficking machinery which reflected by the higher abundance of the corresponded proteins listed in Table 13.

Hyphal tip growth depends on the coordination of vesicle transport using actin and other microtubule cytoskeletons such as kinesin which was shown to be upregulated in the proteome level upon *imp* overexpression. Hyphal growth does not only depend on polarized secretion but also needs endocytosis. This evidence shows that the recycling of the membrane and vesicles sorting is necessary to accelerate hyphal tubes elongation. Two putative vacuolar sorting proteins VPS5/SNX1 and VPS1/dynamin were induced more than two-folds in *imp* overexpressing mutant (Table 13 and S6). Growth of filamentous fungal hyphae is controlled by exocytosis at the tip of the hyphae and cytoplasmic-based forces that push the cytoplasm towards the apical flexible cell wall, as it is proposed in the Figure 34. The membrane protein t-SNARE is involved in endocytosis as well as exocytosis and localization of proteins that are cycled by the late Golgi apparatus compartment. SNF7, VPS1, and VPS5 are small coil-coiled proteins required for the function, structure and formation of multivesicular body and important for cellular vacuolization and apoptosis. Nuclear transport factor 2 is known to interact with the small GTPase Ran protein that is pivotal in the nucleocytoplasmic transport occupying filamentous actin in the process of nuclear transportation. On the other hand, SRP is needed for protein translocation into the endoplasmic reticulum (ER). After proteins are translated, they must reside in the appropriate localization or must be transported outside of the cell such as secreted protein. Protein translocation takes place either during translation or after translation and this occurrence depends on the signal recognition particle (SRP) interacting with specific receptor.

**Table 13. List of membrane trafficking related proteins in OEIMP4 against EVC1 strain that could be involved in fungal endo- and exocytosis.**

| JGI Schco3 ID | Folds | Go Desc molecular function | Go Desc Biological Process | Interpro Desc | Cellular component | KOG Group | KOG Class | KOG Desc |
|---|---|---|---|---|---|---|---|---|
| 2598708 | 2.89 | actin binding | | Actin-binding, cofilin/tropomyosin type | intracellular | Cellular processes and signaling | Cytoskeleton | Drebrins and related actin binding proteins |
| 2541047 | 2.69 | microtubule motor activity,ATP binding | microtubule-based movement | Kinesin, motor region | microtubule associated complex | Cellular processes and signaling | Cytoskeleton | Kinesin-like protein |
| 2674409 | 2.33 | | | | | Cellular processes and signaling | Cytoskeleton | Predicted actin-bundling protein |
| 2681330 | 2.31 | catalytic activity, small protein activating enzyme activity, binding, oxidoreductase activity | protein modification, ubiquitin cycle, metabolism | UBA/THIF-type NAD/FAD binding fold | | Cellular processes and signaling | Cytoskeleton | Predicted actin-bundling protein |
| 2704529 | 23.80 | 7S RNA binding | SRP-dependent cotranslational membrane targeting | Signal recognition particle, SRP19 subunit | | Cellular processes and signaling | Intracellular trafficking, secretion, and vesicular transport | Signal recognition particle, subunit Srp19 |
| 2262522 | 23.70 | | protein transport | Snf7 | | Cellular processes and signaling | Intracellular trafficking, secretion, and vesicular transport | Protein involved in glucose derepression and pre-vacuolar endosome protein sorting |
| 2603193 | 21.68 | ATPase activity, coupled to transmembrane movement of substances, ATP binding | transport | ABC transporter, transmembrane region | integral to membrane | Cellular processes and signaling | Intracellular trafficking, secretion, and vesicular transport | Mitochondrial Fe/S cluster exporter, ABC superfamily |
| 2597192 | 21.66 | DNA binding | DNA methylation | C-5 cytosine-specific DNA methylase | | Cellular processes and signaling | Intracellular trafficking, secretion, | Golgi transport complex subunit |

| | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| | | | | | | | and vesicular transport | |
| 2678633 | 4.27 | acyltransferase activity, oxidoreductase activity | | Acyltransferase ChoActase/COT/CPT; Aldo/keto reductase | | Cellular processes and signaling | Intracellular trafficking, secretion, and vesicular transport | Vesicle coat complex AP-3, beta subunit |
| 2693248 | 3.49 | 7S RNA binding | SRP-dependent cotranslational membrane targeting | Signal recognition particle, SRP72 subunit, RNA-binding | | Cellular processes and signaling | Intracellular trafficking, secretion, and vesicular transport | Signal recognition particle, subunit Srp72 |
| 2460397 | 3.00 | | protein-mitochondrial targeting, mitochondrial inner membrane protein import | Zinc finger | Mitochondrial intermembrane space protein transporter complex | Cellular processes and signaling | Intracellular trafficking, secretion, and vesicular transport | Mitochondrial import inner membrane translocase, subunit TIM8 |
| 2700443 | 2.96 | binding, ARF guanyl-nucleotide exchange factor activity | | Armadillo-type fold, SEC7-like | intracellular | Cellular processes and signaling | Intracellular trafficking, secretion, and vesicular transport | Guanine nucleotide exchange factor |
| 1185145 | 2.44 | protein transport | | Snf7 | | Cellular processes and signaling | Intracellular trafficking, secretion, and vesicular transport | Vacuolar assembly/sorting protein DID2 |
| 2698910 | 2.24 | protein binding, serine-type peptidase activity | cell communication, proteolysis and peptidolysis | Phox-like | | Cellular processes and signaling | Intracellular trafficking, secretion, and vesicular transport | Membrane coat complex Retromer, subunit VPS5/SNX1, Sorting nexins, and related PX domain-containing proteins |
| 2678145 | 2.14 | DNA binding, transcription factor activity | regulation of transcription, DNA-dependent | DNA-binding | | Cellular processes and signaling | Intracellular trafficking, secretion, and vesicular transport | Cysteine protease required for autophagy - Apg4p/Aut2p |
| 2007874 | 2.14 | | transport | Nuclear transport factor 2 | intracellular | Cellular processes and signaling | Intracellular trafficking, secretion, and vesicular transport | Nuclear transport factor 2 |
| 2595664 | 2.12 | protein binding | vesicle-mediated transport | t-SNARE | membrane | Cellular processes and signaling | Intracellular trafficking, secretion, and vesicular transport | SNARE protein Syntaxin 1 and related proteins |
| 2573839 | 2.22 | GTPase activity, GTP binding | | Dynamin, GTPase region | | Poorly characterized | General function prediction only | Vacuolar sorting protein VPS1, dynamin, and related proteins |

## 3.12 The effect of Brefeldin A on membrane trafficking in *imp* overexpressing mutants

Fungal endocytosis and exocytosis are the key processes of hyphal growth which are known to be stimulated by the overexpressing of *imp*. Overexpression of *imp* mutants and control strains were treated with Brefeldin A (BFA). BFA is known as a potent inhibitor of protein secretion in eukaryotic cells including filamentous fungi. BFA initiates dramatic effects on the structure and function of intracellular organelles, particularly the Golgi apparatus. From the result. we could observe that BFA alters the growth of control strains and *imp* overexpressing mutants. Control strains grow slower than *imp* overexpressing mutants in the presence of 50 µg/ ml BFA (Figure 35).

Therefore, we examined further the effect of BFA on the synaptic vesicles and vacuoles under Laser Scanning Microscope (LSM). From the microscopic evaluation. BFA affects the size of the vacuole in the control strains. Vacuole size in the BFA treated T33 and empty vector control (EVC1) were bigger than in the control without BFA (Figure 35 and 36). Occasionally, wavy hyphae in the T33 without addition of BFA were observed, but this BFA effect was not shown in EVC1 strain.

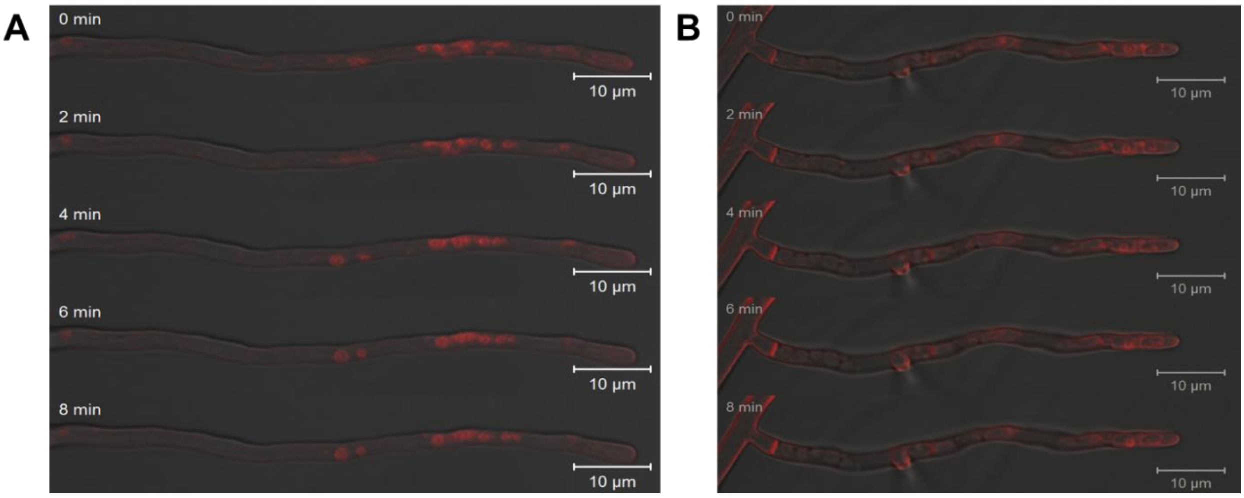


Figure 35. Parental strain T33 without BFA (A) and parental strain T33 with 50 µg/ ml BFA (B), stained by SynaptoRed FM4-64 monitored with time lapse mode for 8 min.

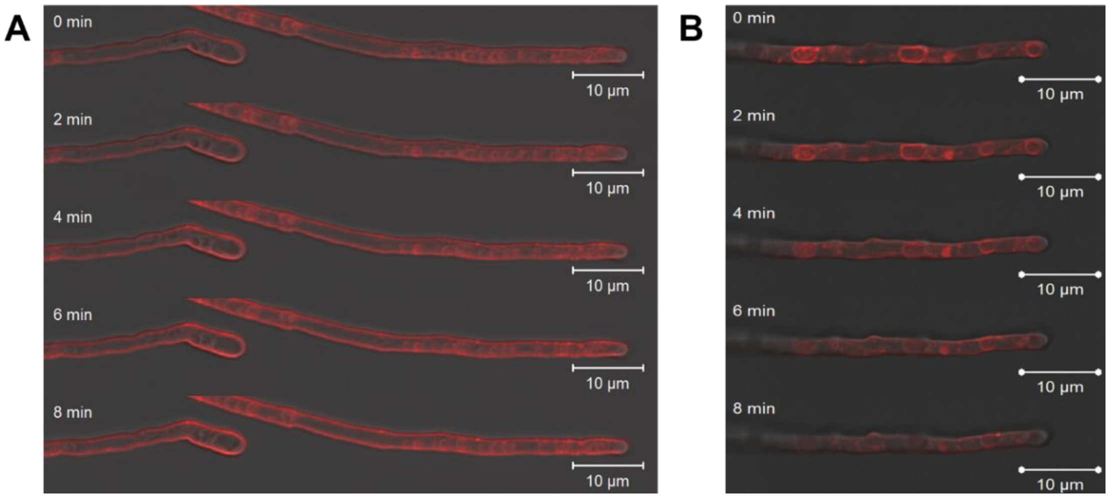


Figure 36. Control strain EVC1 without BFA (A) and control strain EVC1 with 50 µg/ ml BFA (B), stained by SynaptoRed FM4-64 monitored with time lapse mode for 8 min.

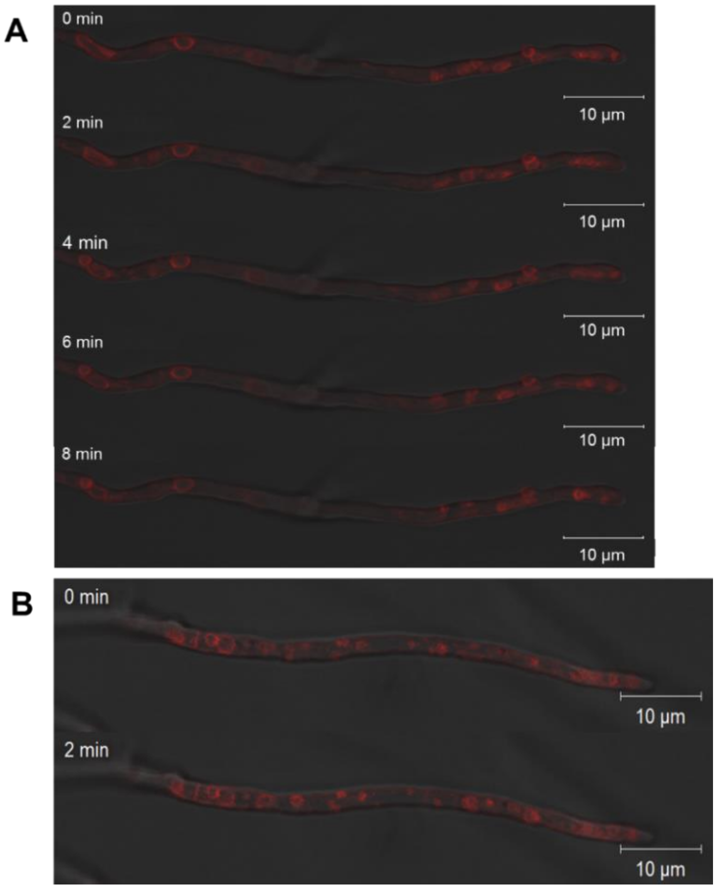


Figure 37. OEIMP4 strain without BFA (A) and OEIMP4 strain with 50 µg/ ml BFA (B), stained by SynaptoRed FM4-64 monitored with time lapse mode for 8 min.

On the other hand, the *imp* overexpressing mutants showed the typical large vacuoles which are more abundant in comparison to both control strains. BFA treated *imp* overexpressing mutants possess similar vacuoles type. The results of live-cell imaging experiments were consistent with the previous reports in proteome analysis showing that cellular trafficking machinery in the *imp* overexpressing mutants are higher in abundance. For instance, Snf72 protein that is responsible for vacuolar assembly and sorting protein is induced, therefore the large vacuoles were shown in both *imp* overexpressing strains. Other protein such as t-SNARE which facilitate Golgi to pre-vacuolar compartment transport and dynamin that is responsible for clathrin-dependent endocytosis and other vesicular trafficking processes, might reflect the large vacuoles that are generated by the *imp* overexpressing mutants OEIMP4 and OEIMP6 (Figure 37 and 38). BFA did not affect the vacuolar trafficking and did not alter the vacuole morphology compared to the *imp* overexpressing mutants without addition of BFA.

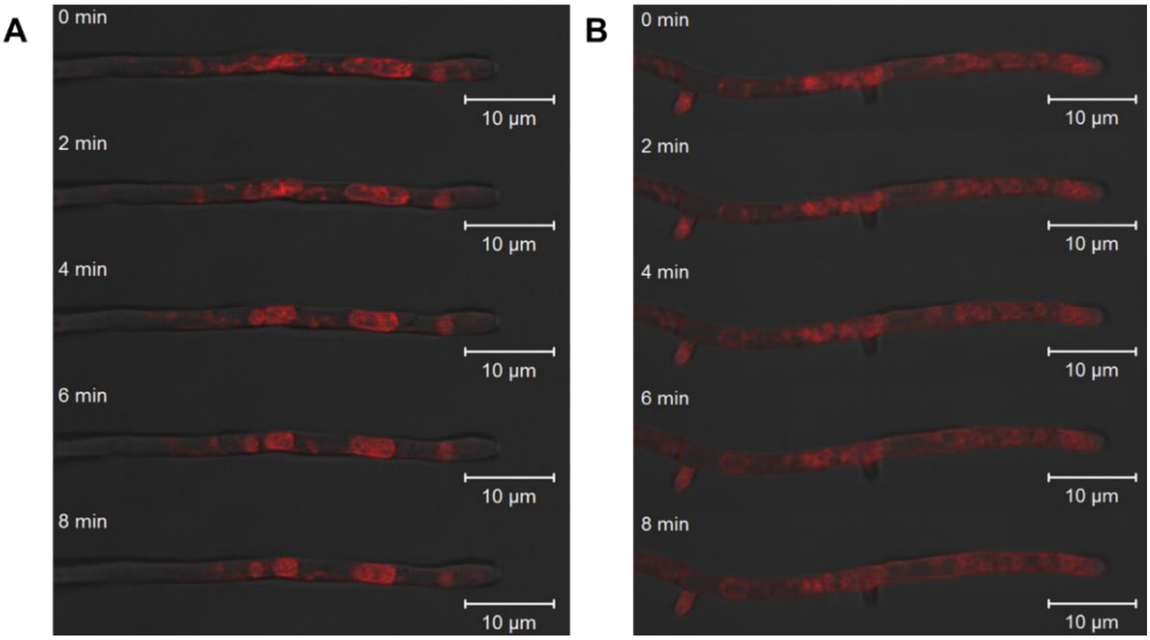


Figure 38. OEIMP6 strain without BFA (A) and OEIMP6 strain with 50 µg/ ml BFA (B), stained by SynaptoRed FM4-64 monitored with time lapse mode for 8 min.

### 3.13 Inositol monophosphatase in developmental stages of *S. commune*

Inositol monophosphatase gene expression was analyzed during mating. Timed samples were obtained starting from 0 hours - 96 hours. The gene expression of *imp* is stable during mating for four days (Figure 39), indicating that inositol monophosphatase is not involved during mating. The *imp* mRNA level expression after 24 hours mating is not altered. There is down-regulation of *imp* gene expression after 48 hours and 96 hours mating, although it is considered as insignificant lower expression. On the other hand, *imp* gene expression was altered from one stage to the other during mushroom development, consistent with the gene expression analysis from the previous study by Ohm (2010). In dikaryon, primordia, and fruiting body, *imp* gene expression is decreased in comparison to the monokaryon. The repression of *imp* gene expression in dikaryon is remarkable at 32.6-folds. This evidence showed that in dikaryon, inositol as the production of inositol monophosphate dephosphorylation by inositol monophosphatase, is not required, therefore it is less expressed in dikaryon. In primordia and fruiting bodies, *imp* gene expression is down-regulated within 3.76-folds and 4.36-folds in comparison to monokaryotic mycelia, respectively. In the proteome analysis, inositol monophosphatase did not show as a regulated protein during sexual development, but other proteins involved in phosphatidylinositol (PI) signaling pathway such as phosphatidylinositol 3- and 4- kinase and myo-inositol 1- phosphate synthase were repressed. Both proteins were repressed in the fruiting bodies, additionally phosphatidylinositol 3- and 4- kinase was found to be repressed in dikaryon and primordia as well.

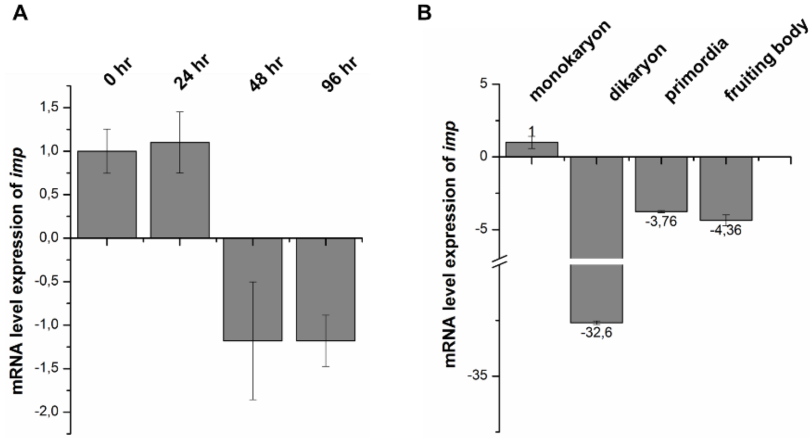


Figure 39. Inositol monophosphatase gene expression during mating of *S. commune* (A) and in different developmental stages of *S. commune* (B).

### 3.14 Putative crosstalk between Ras1 and Imp

The previous microarray study showed that inositol monophosphatase was repressed in the transcriptomic level upon Ras activation (Knabe *et al.*, 2013). In this study, the repression of *imp* gene was confirmed via RT-qPCR. In addition, the knock-out of *rasgap1* mutant was included in the gene expression analysis. As it has been known that GAP1 is the negative regulator for Ras1, the deletion of *rasgap1* leads to the constitutional activation of Ras1, giving the similar effect with the active Ras1 mutant *ras1*$^{G12V}$. RT-qPCR experiment gave a result indicating a cross-talk between Ras1 and inositol monophosphatase (Figure 40 B), as the *imp* gene was repressed more than 50-folds in *ras1*$^{G12V}$ and 34-folds in Δ*ras1gap*. The addition of LiCl showed a different effect in both Ras1 active mutants. Lithium treated *ras1*$^{G12V}$ showed a further remarkable repression of *imp* gene expression up to 75-folds and on the other hand the lithium treated Δ*ras1gap* shows the lower repression of *imp* gene expression in comparison to the non-treated Δ*ras1gap* which was down-regulated within 20-folds. Furthermore, inositol monophosphatase activity in all strains with or without LiCl presence was examined and the result was correlated with the gene expression analysis. Interestingly, the inositol monophosphatase activity in the Ras1 active mutants are correlated with the *imp* gene expression analysis. Inositol monophosphatase activity are reduced upon Ras1 activation. which are observed in all Ras1 active strains. Activity of inositol monophosphatase were reduced up to sixty-five percent in the *ras1*$^{G12V}$ and drastically reduced in the Δ*ras1gap* (up to

97 % reduction). In the wild-type, lithium reduced the activity of inositol monophosphatase within thirty-one percent.

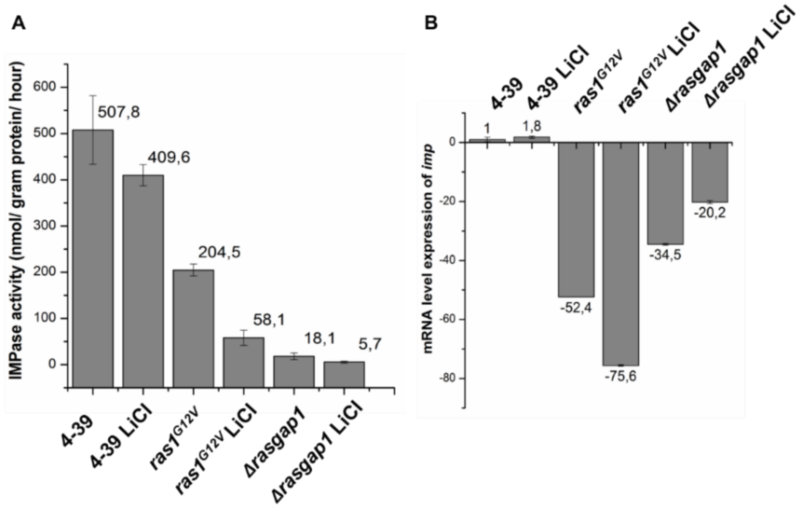


Figure 40. Inositol monophosphatase activity of wild type 4-39. Ras1 active mutants *ras1*$^{G12V}$ and Δ*rasgap1* in the presence of LiCl and without LiCl (A) and mRNA level expression of *imp* with and without LiCl (B).

### 3.14.1 Ras1 activation alters inositol phosphates profile in the presence of lithium

Inositol phosphates are formed rapidly as a result of phosphatidylinositol – phospholipase C (PI-PLC) activity. The most famous molecule inositol 1,4,5-trisphosphate and its succeeding metabolites showed different profile under lithium treatment in wild type 4-39. In contrast to wild type, there is no significant difference between the lithium treated and non-lithium treated *ras*$^{1G12V}$ mutant. In wild type 4-39, an astonishing difference was observed in inositol phosphate profile, from the inositol monophosphate until the inositol pyrophosphates level. In wild type, it is unknown or even unlikely, that the lithium target directly the inositol phosphate kinases such as inositol multiphosphate kinases (Imk), inositol pentakisphosphate kinase (Ipk), and inositol polyphosphate kinase (Kcs1 and Vip1). The reduction of inositol monophosphate level in the wild type suggests that lithium might target also the enzyme that dephosphorylate $IP_2$ to the inositol monophosphate, thus there is a tail-back phenomenon, where the $IP_3$ was pushed to be phosphorylated to the higher inositol phosphate species.

This explains that lithium also might affect indirectly the cellular activity of inositol phosphate kinases that plays role in inositol phosphate phosphorylation. This phenomenon does not occur in the Ras1 active mutant, indicating that Ras1 activation prevents the effect of lithium to produce higher level of succeeding inositol phosphates from $IP_3$, thus the phosphorylation of inositol triphosphate ($IP_3$) to the following higher inositol phosphates is retained in certain level. Furthermore, in the Ras1 active mutant the inositol monophosphate level was not affected by lithium. It can be concluded that Ras1 activation prevents the attenuation of inositol monophosphatase and also inositol bisphosphatase/ inositol polyphosphate phosphatase by lithium, therefore prevents the channeling of inositol phosphates towards the inositol phosphate kinases activity.

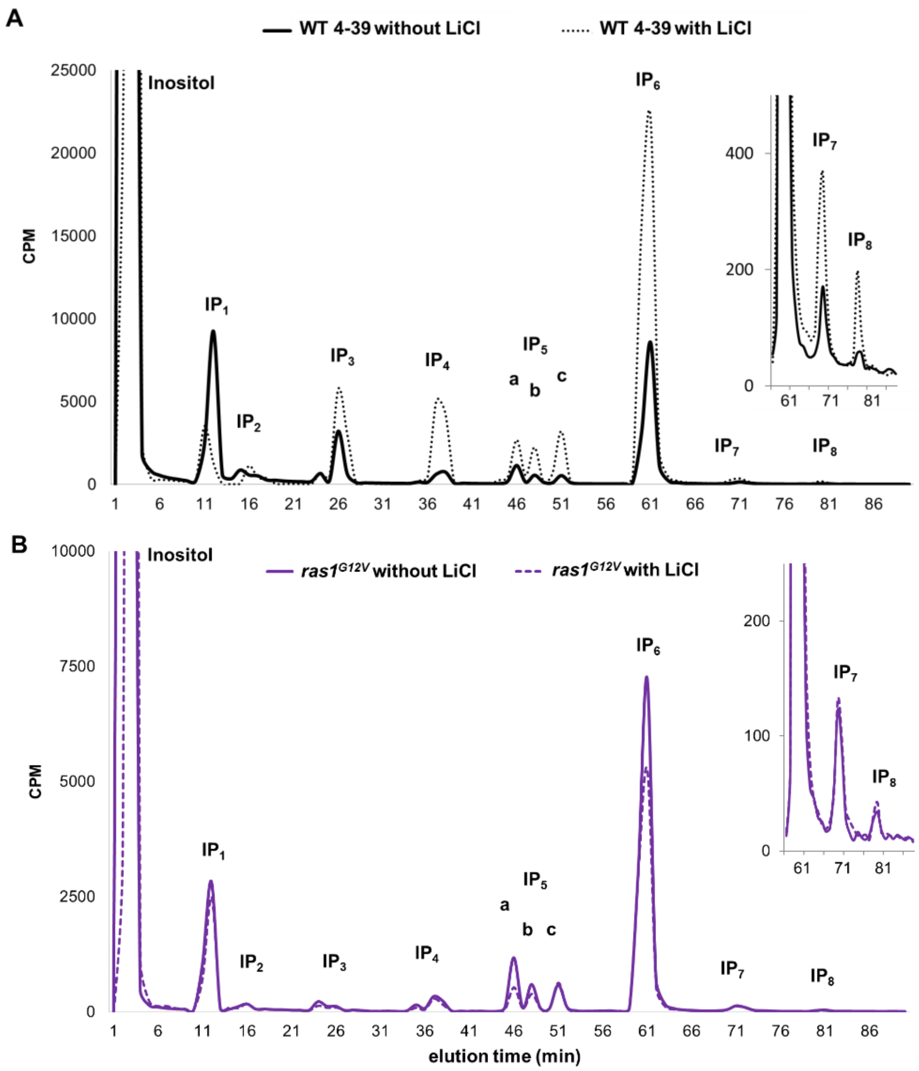


Figure 41. Inositol phosphate profile in wild type 4-39 (A) and $ras^{1G12V}$ mutant (B) in the presence (represented in a dotted line) or absence (depicted in a smooth line) of LiCl for 24 hours. A remarkable elevation on the inositol phosphate profile of the wild type did not occur when Ras1 is activated.

Hyper-synthesizing of phosphorylated inositol phosphate species ($IP_3$-$IP_8$) that was found in wild type, did not occur in Ras1 active mutant (Fig 41 A and B). It is important to mention that the growth of the $ras1^{G12V}$ was reduced although not significant in comparison to the wild type 4-39. The cultivation was executed with the same amount of starting material, however $ras1^{G12V}$ had slightly less growth than wild-type 4-39 after 24 hours grown in liquid MM-U. Lithium induces stress in hyphae and represses growth significantly in *S. commune* as it was examined also on CYM plates (Figure S5).

The difference is, that the growth of wild-type 4-39 was more susceptible to the lithium than Ras1 active mutant $ras1^{G12V}$. Because of this reason, inositol phosphate profile of both strains could not be compared directly. In principle, inositol phosphate level executed by the *ras1*$^{G12V}$ was considerably lower than the inositol phosphate level from the wild type 4-39.

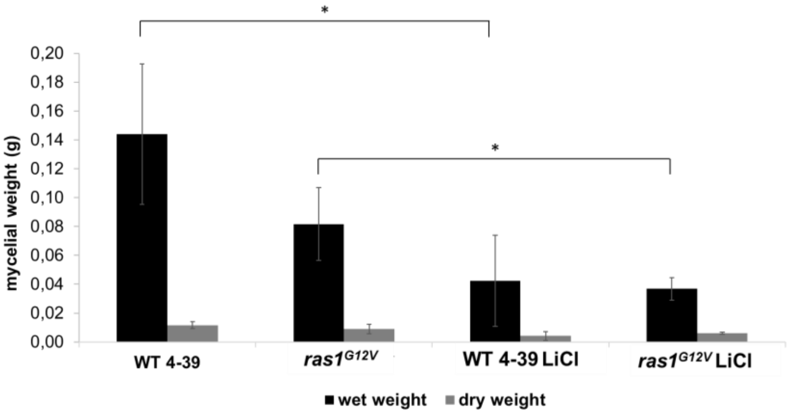


Figure 42. Dry or wet mycelial weight reflects the growth rate of *S. commune* WT 4-39 and *ras1*$^{G12V}$.

### 3.14.2 Gene expression of *ras1* and *ras2* prior to *imp* overexpression

The alteration of inositol phosphate profile by Ras1 activation prompted us to check the transcriptomic level of *ras1* and *ras2* gene in the *imp* overexpressing mutants. Gene expression of *ras1* is induced within 3.9-folds and 2.4-folds in the *imp* overexpressing mutants OEIMP4 and OEIMP6, respectively.

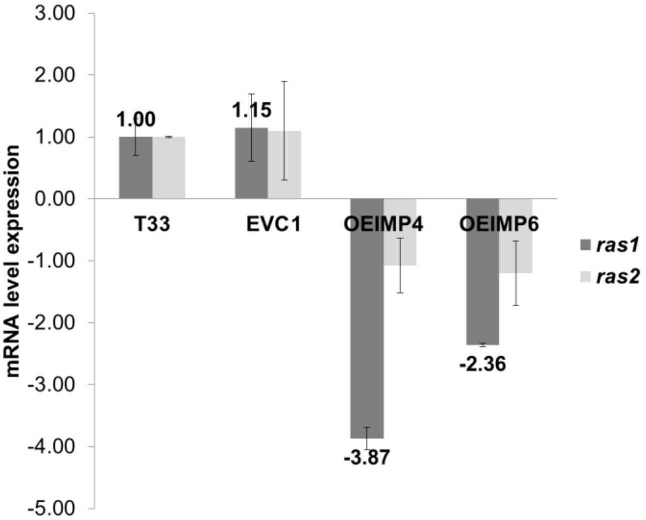


Figure 43. Alteration of *ras1* and *ras2* gene expression in *imp* overexpression OEIMP4 and OEIMP6 mutants via RT-qPCR. The graph shows a relevant information about the induction of *ras1* gene expression, but not *ras2* gene expression prior to *imp* overexpression.

On the other hand, the *ras2* mRNA level was not altered in both *imp* overexpressing mutants. This outcome clarifies the evidence of cross-talk between Ras signaling and inositol phosphate signaling specifically through Ras1 and Imp. Presumably, the interaction could be done through another protein such as phosphatidylinositol 3-kinase (PI3-kinase/ PI3K), since PI3K is able to bind to Ras. If it is so, the alteration of inositol monophosphatase gene regulation and activity that affect inositol phosphates metabolism is only secondary effect. This assumption should be proven by some further investigations. Additionally, the role of Ras2 in *S. commune* was never identified, on the other hand, the role of Ras1 is to mediate polar growth and morphogenesis.

### 3.14.3 Mating and fruiting bodies of Ras1 active mutants and *imp* overexpressing mutants

Ras1 activation leads to the unilateral mating and failure in sporulation (Knabe *et al.*, 2013). From the results shown above, Ras1 seems to be a negative modulator for inositol phosphate metabolism by repressing inositol monophosphatase. It was hypothesized that normal fruiting body formation can be complimented with the inositol monophosphatase overexpression. Mating was established between the Ras1 active mutant and *imp* overexpressing mutant. The dikaryotic hyphae could be observed in all cases of the mating with Δ*rasgap1* and *ras1*$^{G12V}$ after 3-5 days inoculation, right after mycelia from two hyphae fused. Clamp cells look normal and binucleated cells could be observed in all cases, which indicate that the mating between Ras1 active mutant and *imp* overexpressing mutants and all control strains are successful.

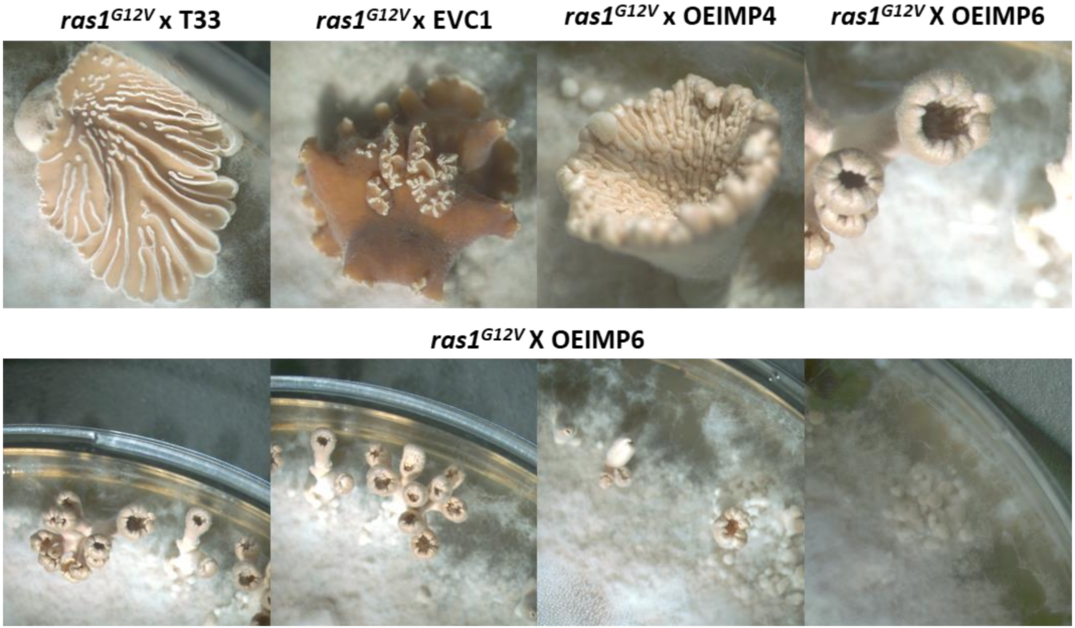


Figure 44. Fruiting bodies produced by the mating interaction between *ras1*$^{G12V}$ and *imp* overexpressing mutant OEIMP4 and OEIMP6 along with the control strains (T33 as parental strain and EVC1 as the empty vector control strain) on MMUT Trp 0.6 mM.

Fruiting bodies which were produced from the mating plates between *ras1*$^{G12V}$ mutant and *imp* overexpressing mutants appeared after 28-30 days inoculation. The fruiting body which was produced in the plate *ras1*$^{G12V}$ crossed with T33, resulted in a normal fruiting body that has a well-developed lamella and normal basidiospores (Figure 44). Surprisingly, a defect fruiting body with a peculiar, non-developed lamella and lacking basidiospores was produced from the plate of *ras1*$^{G12V}$ crossed with EVC1. The unusual feature of closed-cup fruiting bodies was observed in the plate of *ras1*$^{G12V}$ crossed with OEIMP4 and OEIMP6. Thicker lamellae were shown in the surface of fruiting body and the fruiting body lacks basidiospores. High number of fruiting bodies were produced in the plate which *ras1*$^{G12V}$ crossed with OEIMP6, which was not shown in the plate of *ras1*$^{G12V}$ crossed with OEIMP4.

Mating between *ras1*$^{G12V}$ and *imp* overexpressing mutants showed unclear result (Figure 45), therefore it is hard to conclude the outcome from this investigation. Unsystematic result was also found in the mating plate between Δ*rasgap* and two *imp* overexpressing mutants. After 18 days post inoculation, the primordia of mating interaction plate between Δ*rasgap1* and EVC1 and T33 appeared. In mating plate with the *imp* overexpressing mutants, the primordia appeared on the 20-21 days post inoculation. All fruiting bodies could be observed after 24 days post inoculation. The morphology of fruiting body in different mating plates could not be distinguished. The only difference between the mating with *imp* overexpressing mutants and the control strains, are the amount of fruiting bodies produced. More fruiting bodies produced was shown in the mating plates of control strains compared to mating plates with *imp* overexpressing mutants. It was also noticed that the fruiting bodies produced from the mating with T33 has thicker basidiocarp rather than the one which was crossed with the other strains. In some cases, it was observed that fruiting bodies from the mating plate Δ*rasgap* x OEIMP4 have no gills (Figure 45).

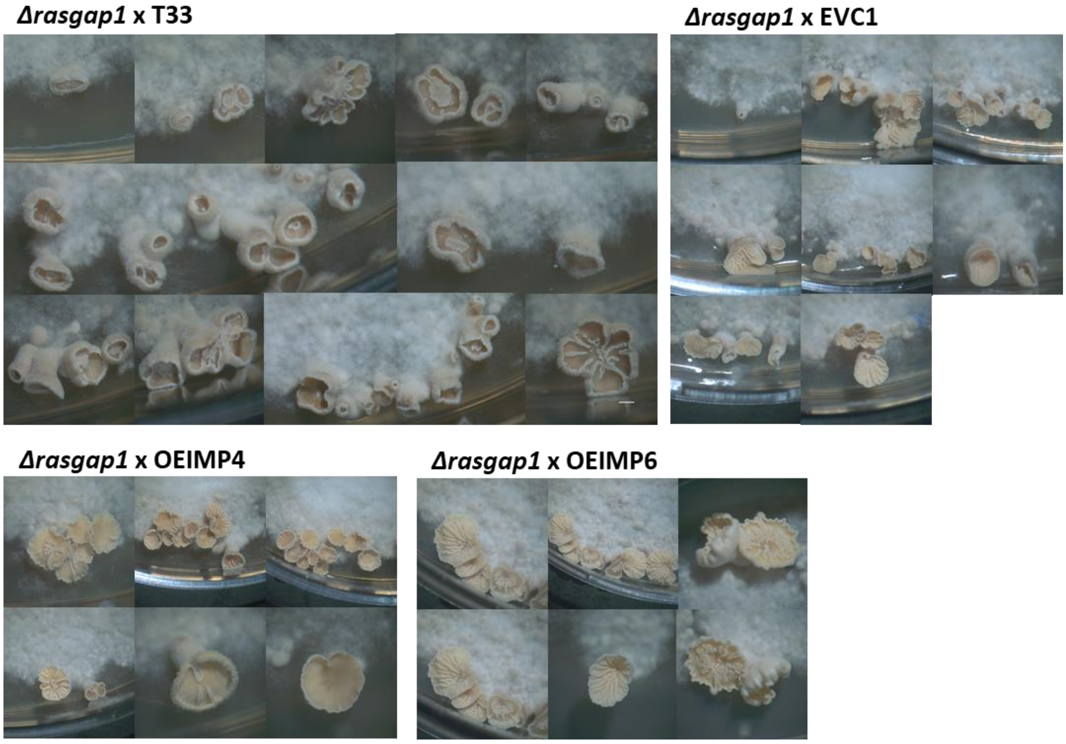

**Δ*rasgap1* x T33**
**Δ*rasgap1* x EVC1**
**Δ*rasgap1* x OEIMP4**
**Δ*rasgap1* x OEIMP6**

Figure 45. Different fruiting body morphology as results of mating interaction between Δ*rasgap* and two *imp* overexpressing mutants OEIMP4 and OEIMP6 on MM-UT (0.6 mM Tryptophan). Lack of developed gill is the most striking feature of fruiting bodies that only executed by *imp* overexpressing mutant OEIMP4 crossed with Δ*rasgap*. Fruiting body produced by mating interaction with the parental strain T33 possess thick lamellae. This feature is missing in EVC1 crossed with of *imp* overexpressing mutants.

# 4 Discussion

## 4.1 Inositol as a building block for inositol phosphate and phosphoinositide

Inositol-based regulatory systems are omnipresent across most life forms especially in eukaryotes from the unicellular yeast, metazoan, fungi, plants, and mammalian. They are involved in multifaceted cellular processes, including signaling from membrane cell-surface receptors, embryonic development, chemotaxis, transcription control, mRNA export, DNA repair, vesicle trafficking, and homeostasis of phosphates. In filamentous fungi, inositol phosphate has been investigating only in the ascomycete *Podospora anserina* and to the best of my knowledge it was never investigated in basidiomycete mushroom forming fungi before this study. Inositol phosphate is a water-soluble molecule that consists of inositol as the building block and one or more phosphate that can attach to the inositol backbone, replacing six possible hydroxyls (the 2-hydroxyl is axial, and the other five hydroxyls are equatorial). Inositol is preferred since it is chemically stable than other sugar moieties such as glucose, fructose, or ribose (Saiardi, 2017). There are more than 60 possible inositol phosphates that potentially can be further expanded by adding the pyrophosphate molecules instead of monophosphate, for instance the inositol pyrophosphates 5-diphosphoinositol-pentakisphosphate ($IP_7$) and 5-Bis-diphosphoinositol-tetrakisphosphate ($IP_8$) (Irvine and Schell, 2001). The metabolism of this class of molecules is vastly dynamic and the enzymes that are involved for their metabolism are evolutionary conserved among organisms (Saiardi, 2012a). Not only as the base molecule for inositol phosphates, inositol is also the precursor of all inositol-phospholipid containing compounds so-called phosphoinositide. Phosphoinositide is the key signaling molecules in the membrane cell-surface receptors. Phosphoinositide as its name, is similar to inositol but in addition they possess a hydrophobic component diacylglycerol moiety containing two fatty acids esterified onto glycerol group, that is in turn attached by the diester phosphate to 1-hydroxyl group of the inositol backbone. Since they contain the hydrophobic component, therefore they are not water-soluble (Irvine and Schell, 2001). Seven phosphoinositide species can be produced through interactions which is facilitated by their headgroups (Di Paolo and De Camilli, 2006). Phosphoinositide facilitates high range responses and furthermore act as constitutive membrane signals including modulation of membrane trafficking, the cytoskeleton polymerization, nuclear events, and the transport functions of membranes-related processes. The origin of the inositol phosphate metabolism comes from the most prominent phosphoinositide molecule phosphatidylinositol 4,5-bisphosphate ($PIP_2$). The birth of inositol phosphate starts once the phosphatidylinositol-phospholipase C (PI-PLC) cleaves $PIP_2$ into diacylglycerol (DAG) and the second messenger molecule inositol 1,4,5-trisphosphates ($IP_3$) (Berridge, 2009). $IP_3$ binds to $IP_3$ receptors resided in the endoplasmic reticulum membrane and regulates calcium release into the cytosol and this occurrence triggers numerous cellular processes. Phosphorylation and dephosphorylation of $IP_3$ molecules are performed by the group of inositol phosphate phosphatases and inositol phosphate kinases. The turnover of phosphoinositide or inositol phosphate molecules to another are highly connected to wide range of cellular processes, thus making the phosphatidylinositol signaling and inositol phosphate metabolism are crucial to the living organisms. This study is only focused in the water-soluble inositol phosphate. *In silico* analysis revealed the inositol phosphate metabolism of basidiomycete mushroom forming fungus *S. commune* along with the enzyme that generates the multivariant of inositol phosphates molecule.

### 4.1.1 Inositol phosphate metabolism in *S. commune*

Inositol phosphates metabolism in *S. commune* was revealed by *in silico* analysis and compared to other organisms such as human and yeast *S. cerevisiae*. In human and *S. cerevisiae*, metabolism of inositol phosphate is well identified. Biochemical measurement and genomic data on the inositol phosphate metabolism in *S. commune* are combined to have a complete analysis. According to the analysis of sequence similarity between human and *S. commune* as well as *S. cerevisiae* and *S. commune*, *S. commune* possesses two inositol phosphate phosphatases which are inositol monophosphatase and inositol 1,4,5-trisphosphate 5-phosphatase. Inositol monophosphatase generates inositol by dephosphorylating inositol 4-P and Inositol 1,4,5-trisphosphate 5-phosphatase works on dephosphorylation from inositol 1,3,4,5-$P_4$ to inositol 1,3,4-$P_3$ and dephosphorylation from inositol 1,4,5-$P_4$ to inositol 1,4-$P_2$. Human and *S. cerevisiae* possesses more than one inositol monophosphatase while *S. commune* has only one inositol monophosphatase. In addition, *S. commune* has inositol polyphosphate 5-phosphatase, possibly that works on dephosphorylation of any inositol 4,5-bisphosphates and inositol 1,5-bisphosphates to inositol 4-monophosphate and inositol 1-monophosphate. Multiple inositol polyphosphate phosphatase is also found in the *S. commune* genome, presumably it dephosphorylates any high species of inositol phosphate that possesses histidine acid phosphatase (HAP) domain.

It has been revealed that *S. commune* has three inositol phosphate kinases including inositol polyphosphate kinase Kcs1/ inositol-hexakisphosphate 5-kinase, inositol polyphosphate multikinase, and inositol pentakisphosphate 2-kinase. Inositol polyphosphate kinase Kcs1/ inositol-hexakisphosphate 5-kinase is responsible for phosphorylation from inositol 1,2,3,4,5,6-hexakisphosphate to 5-diphosphoinositol-pentakisphosphate and phosphorylation from 1-diphosphoinositol-pentakisphosphate to bis-diphosphoinositol-tetrakisphosphate. Inositol polyphosphate multikinases (Imk) is responsible for phosphorylation from inositol 1,3,4,5-tetrakisphosphate to inositol 1,3,4,5,6-pentakisphosphate, phosphorylation from inositol 1,4,5-trisphosphate to inositol 1,4,5,6-tetrakisphosphate, phosphorylation from inositol 1,4,5,6-tetrakisphosphate to inositol 1,3,4,5,6-pentakisphosphate, and phosphorylation from inositol 1,3,4,6-tetrakisphosphate to inositol 1,3,4,5,6-pentakisphosphate. Inositol pentakisphosphate 2-kinase (Ipk) works on phosphorylation from inositol 1,3,4,5,6-pentakisphosphate to inositol 1,2,3,4,5,6-hexakisphosphate. Ipk can be found in human and *S. commune* but it does not exist in *S. cerevisiae*. In *S. commune*, $IP_6$ can be generated from two different inositol phosphate kinases. This hypothesis must be confirmed by knock-down experiment of Ipk and Imk, since deletion will not be a suitable approach considering these two genes encoding these proteins are essential. In comparison to fungi, human has more complex inositol metabolism. In addition to mentioned phosphatases and kinases, human possesses three inositol-trisphosphate 3-kinases, three inositol polyphosphate 5-phosphatases, inositol-tetrakisphosphate 1-kinase, two inositol polyphosphate 4-phosphatase, one inositol polyphosphate 1-phosphatase, and three inositol polyphosphate 5-phosphatases. Presumably, Imk of *S. commune* is the substitution enzymes for inositol-trisphosphate 3-kinase and inositol-tetrakisphosphate 1-kinase.

To study inositol phosphate in *S. commune*, cells were labelled with tritium [$^{3}$H] inositol for 24 hours. Other alternatives for inositol labelling are with radioactive orthophosphate [$^{32}$P] or [$^{32}$P] and also carbon [$^{14}$C]. Tritium [$^{3}$H] was chosen since its emitted electron has relatively low energy and thus it can travel only a few millimeters in the air and it is not harmful since it is not able to diffuse into the skin (Wilson and Saiardi, 2017), therefore it is safer and handy to work with. Inositol labelling with tritium [$^{3}$H] in *S. commune* appeared to be successful. Inositol phosphates profile of *S. commune* shows that this fungus is able to produce the multivariant of inositol phosphates from inositol monophosphate ($IP_1$) to the inositol pyrophosphates ($IP_7$ and $IP_8$), thus creating a huge metabolic network of inositol phosphates. The convolution of the inositol phosphate metabolic network with its high number of metabolites, allowing to consider that inositol phosphates other than the famous second messenger $IP_3$ have also important cellular functions.

In *S. commune*, it was shown that $IP_6$ is the most abundant inositol polyphosphate. A previous report was also shown in other organisms such as slime molds *Dictyostellium discoideum* (Pisani *et al.*, 2014) and plants (Raboy, 2001). Inositol hexakisphosphate ($IP_6$) is the most predominant form of phosphorylated inositol in the eukaryotic cell. The significance of Ins 1,3,4,5,6-$P_5$ and $IP_6$ has also been found in all mammalian cells in greater amounts in comparison to any other inositol polyphosphates (Menniti *et al.*, 1993). $IP_6$ or also called phytate has been found accumulated in seed plants and has been known to be the most abundant molecule in seed. In the seeds, $IP_6$ normally consists of less than one percent of the seed dry weight and this amount is responsible for two-thirds of total seed phosphorus (Raboy, 2001). In *S. commune*, $IP_3$ dephosphorylates to the lower inositol phosphate species $IP_2$ and $IP_1$ then inositol is generated. $IP_3$ also phosphorylates to $IP_4$, $IP_5$, $IP_6$, and further to high energy inositol pyrophosphates $IP_7$ and $IP_8$. Inositol pyrophosphates have been known to govern cell physiology and phosphate homeostasis (Saiardi, 2012b). Inositol phosphates profile in *S. commune* showed that this fungus has the canonical inositol phosphate kinases and inositol phosphatases, that create the complex metabolites of inositol phosphate metabolism.

### 4.1.2 Inositol polyphosphates play role in sexual development of *S. commune*

Many cellular processes alteration is connected with inositol phosphate signaling pathway including developmental changes in organisms such as *Trypanosoma brucei* (Cestari *et al.*,

2018) and *Dictyostellium discoideum* (Luo *et al.*, 2003), and also cell cycle progress of rat thymocytes (Guse *et al.*, 1993). Developmental changes of *T. brucei* correlates with knockdown of inositol polyphosphate multikinase (Imk) (Guse *et al.*, 1993). To analyze the inositol phosphate profile in different sexual development of *S. commune*, inositol phosphate was extracted using perchloric acid after labelling for several hours depending the type of the culture, then neutralized to neutral pH, and analyzed using SAX-HPLC. Astonishingly, there is changes in inositol phosphates associate with the developmental change from monokayrotic haploid mycelia to fruiting bodies, suggesting that inositol phosphates turnover play role in the development of basidiomycete fungus *S. commune*. Fruiting bodies lack $IP_{5a}$ which is known as inositol 1,3,4,5,6-pentakisphosphate, the precursor of 1,2,3,4,5,6-hexakisphosphate generation. For this evidence, it was hypothesized that the inositol phosphate kinases that are responsible for the $IP_5$ and $IP_6$ generation might be altered in gene regulation. Therefore, the gene regulation of two gene candidates that are found similar with human and *S. cerevisiae*, were analyzed. Inositol polyphosphates are generated from the inositol polyphosphate multikinases (Imk) and inositol 2-pentakisphosphate kinase (Ipk) that specifically generates $IP_5$ to $IP_6$. Gene expression of *imk* and *ipk* are not altered in fruiting bodies and it does not correlate with the fact that $IP_6$ level is depleted in fruiting bodies. It is indeed a peculiar fact, that $IP_6$ is less produced in fruiting body and most probably in spores of *S. commune*, since $IP_6$ has been found highly abundant in the zygotic embryo of plants in the form of seeds, grains, and legumes (Sparvoli and Cominelli, 2015). On the other hand, other report showed that Imk plays crucial role in mating and sporulation. Mutation in Imk (previously known as ArgIII) resulted in repression of arginine biosynthesis, induction of arginine catabolism, defect in mating and sporulation (Dubois and Messenguy, 1994). The fact that mRNA level of *imk* is highly down-regulated in primordia of *S. commune*, showed that *imk* might play role in morphogenesis and sexual development of basidiomycete *S. commune*, unfortunately the inositol phosphate analysis in primordia to confirm the involvement of *imk* as in fungal morphogenesis, is still missing. However, the results still suggest that inositol phosphate turnover might be involved in the regulation of fruiting body development.

## 4.2 Lithium effect on inositol phosphate signaling

### 4.2.1 Lithium attenuates inositol monophosphatase and alters inositol phosphates

In this study, it has been shown that lithium has a profound effect on inositol phosphate metabolism in the basidiomycete *S. commune*. The inositol depletion mechanism by lithium is most likely caused by inhibition of inositol monophosphatase and other inositol phosphate phosphatase, in line with the previous studies that showed lithium inhibits inositol monophosphatase activity in *S. cerevisiae* and reduces the expression of the corresponding gene *inm1* (Murray and Greenberg, 1997, 2000). In *S. commune* the significant inhibition of inositol monophosphatase by lithium was observed in the wild type 4-39 concomitant with the slightly reduction of inositol content. The inositol reduction was not high in comparison to the reduced inositol monophosphatase activity because it is assumed that there was a technical problem in mycelial washing. It is presumed that there was some remaining labeled inositol in the medium. Mycelia could not be pelleted and some of the mycelia remained floating in the medium which led to the washing problem and providing excessive labelled inositol in the extraction, which did not come from the hyphae. Nevertheless, the reduction of inositol content could still be observed. Furthermore, inositol uptake from exogenous resources is another reason why the difference between lithium treated and non-treated wild type is not significant (Harwood, 2005). Another study shows that lithium induces alterations in neural activity and changes the development of mammalian, by repressing the inositol supply, the key component for the lipid inositol phosphate signaling phosphatidylinositol by inhibiting the key enzyme that dephosphorylate inositol phosphate to inositol (Berridge and Irvine, 1989). Two enzymatic reactions or pathways known as targets for lithium are inositol monophosphatase and the protein kinase glycogen synthase kinase 3. Lithium reduces the activity of both enzymes through displacing the magnesium as the enzyme cofactor (Brown and Tracy, 2013; Sade *et al.*, 2016). The hypothesis for the inhibitory effects of lithium on enzymatic targets inositol monophosphatase is the metal-binding competition between the inhibitor lithium and the cofactor magnesium on the metal-binding sites. Lithium and magnesium have comparable ionic radii, namely 0.60 and 0.65 Ångström (Å), respectively and both possesses similar physicochemical properties (Dudev and Lim, 2011; Haimovich *et al.*, 2012).

Lithium surprisingly does not only affect the inositol, but also the entire inositol phosphate profile, for instance the reduction can also be observed in $IP_1$ (Figure 17). On the contrary, lithium effect was performed using quantitative analysis of the inositol phosphates in rat's brain by ion chromatography showed elevation of the mass of myo-inositol 1-phosphate and 4-phosphate. however similar effect decrease in content of labeled inositol was observed (Sun *et al.*, 1992). Interestingly lithium induces the production of inositol polyphosphates $IP_3$, $IP_4$, $IP_5$, $IP_6$ as well as the inositol pyrophosphates $IP_7$ and $IP_8$ (Figure 17). Elevated $IP_3$ levels because of lithium have been observed as well in the cerebral cortex of mammalian such as guinea pigs, rabbits, Monkeys, rats (Lee *et al.*, 1992). One possible explanation for elevated inositol polyphosphates and the phosphorylation route of $IP_3$ by kinases is preferred because lithium inhibits the dephosphorylation route of $IP_3$, avoiding the recycle inositol and as a response inducing the entire inositol polyphosphate level of *S. commune*. It indicates that lithium may affect inositol phosphate kinases on molecular level. However, further studies are required to test this hypothesis. According to *in silico* analysis *S. commune* possesses several inositol phosphate kinases including inositol polyphosphate multikinases (Imk) and inositol 2-pentakisphosphate kinase (Ipk) that are responsible to hydrolyze $IP_6$. Inositol polyphosphate kinase Kcs1 and inositol polyphosphate kinase Vip1 act in concert to synthesize the diphosphate group-containing inositol pyrophosphates diphosphoinositol pentakisphosphate (PP-$IP_5$) and bis-diphosphoinositol tetrakisphosphate ($(PP)_2$-$InsP_4$). The elevation of high species inositol phosphates by lithium revealed insights of feedback loop on the attenuation of inositol monophosphatase. Inositol polyphosphate kinases are highly conserved throughout eukaryotes, including higher fungi. It comprises of widely conserved dual domain structures, the N-terminal ATP grasp domain that has putatively kinase activity and the C-terminal histidine acid phosphatase (HAP) domain.

### 4.2.2 Lithium affects fungal growth and sexual development

Lithium has been used for decades to treat bipolar and unipolar affective disorders (Lepkifker *et al.*, 2007), however the mechanism that underlies its therapeutic action is not fully understood. It is important to notice that in previous studies, lithium is performed as an inducer of autophagy in mammalian. Lithium facilitates the allowance of known autophagy substrates and induces autophagy by inhibiting inositol monophosphatase, that resulting in the reduction of free inositol level and level of the most prominent inositol phosphate molecule, the inositol-1.4.5-triphosphate ($IP_3$) levels (Sarkar *et al.*, 2005).

Lithium has been shown to affect negatively the growth rate of the basidiomycete *S. commune*. Lithium lowers significantly the growth concomitantly with the repression of actin cytoskeleton in the proteome level. As it has been always known that actin is involved in hyphal growth, morphogenesis, cytokinesis, and the movement of organelles. Lithium affects the dynamic of actin which was shown under laser scanning microscopy. Perhaps, the reduction of inositol affects the production of Phosphatidylinositol 4,5-bisphosphate ($PIP_2$) and reduced amount of $PIP_2$ may interfere its binding with the actin binding protein, therefore actin regulation might be altered. Morphogenesis in fungal hyphae depends on the localization and the directed transport of vesicles to the hyphal growth site. Hyphal growth is directed with a vesicle supply center which requires actin binding as a cytoskeleton protein. In contrast to our finding, a previous study proved that lithium increases the polymerization of cytoskeletal proteins actin in vitro (Colombo *et al.*, 1991). Another evidence shows that F-actin in *Chlamydomonas flagella* is elongated by lithium with an unknown mechanism (Quarmby, 2014). In line with another fungal study, that lithium mediates the suppression of morphogenesis and growth in fungi yeast form such as *S. cerevisiae* (Masuda *et al.*, 2000) and *Candida albicans* (Martins et al., 2008). On the other hand, the effect lithium on basidiomycete fungal growth has been tested before and the result is in line with the result on this study which show also the defect of fungal growth depending on the lithium concentration given into the media. Mateus *et al.* (2014) insisted that basidiomycete is a group of fungi that more sensitive to the lithium than ascomycetes (Richter *et al.*, 2008). Lithium affect the fungi to alter hyphal diameter and decreased hyphae diameter to reduce the energy needed to produce a cell and fungi might use this energy to adapt for a more suitable environment (Dávila Costa *et al.*, 2011; Mateus *et al.*, 2014). Stress indicator that was shown in hyphal growth of *S. commune* is most probably associated with the induction of stress associated proteins, such as heat shock protein Hsp70 and superoxide dismutase. Changes in environment including elevated temperatures, chemical, and physiological stresses affect the regulation of heat shock proteins (Wu, 1995). Lithium has been known to activate heat shock protein in the inner medullary collecting ducts of rats (Nielsen *et al.*, 2008) and several studies propose that harmful effects of lithium might be related to oxidative stress (Efrati *et al.*, 2005; Kielczykowska *et al.*, 2004). Furthermore, in *S. commune*, induction of some protein related to protein biosynthesis, proteolysis & peptidolysis, and protein modification is observed in lithium treated fungi to show the metabolic response against stress or adaptation to the environmental changes. The fact that superoxide dismutase is increased is interesting and might be linked to the alteration of amino acid metabolism. As it has been known that superoxide dismutase is activated when there is damage to some protein clusters which inactivate key enzymes involved in amino acid and sugar metabolism, thus refers to a mobilization of free iron and stimulation in iron-related toxicity (Culotta, 2001). Lithium treated fungus suffered from repression of some proteins related to the glycolysis metabolism resulting in nutrient limitation, therefore it reflects on the fungal growth defect in the plate. *S. cerevisiae* grown on galactose and treated with relevant concentrations of lithium showed a dramatic drop in flux through glycolysis affecting metabolic shift from fermentation to respiration and induction of glycogen content (Bro *et al.*, 2003). The effect of lithium is similar with the occurrence of nutrient limitation that is shown at the diauxic shift where glucose becomes limited (DeRisi *et al.*, 1997). Group of proteins that are important for citrate (Si)-synthase activity, malate dehydrogenase activity, and glutamine biosynthesis are declined drastically in the presence of lithium. Defect on glutamine biosynthesis might affect signal nitrogen sufficiency since glutamine has been pointed out as a key effector for suppression of nitrogen metabolite (Margelis *et al.*, 2001).

Furthermore, a protein that possesses histidine acid phosphatase (HAP) domain is known to be repressed under lithium treatment. The HAP superfamily is a large functionally group of proteins that share a conserved catalytic subunit centered on a histidine which can be phosphorylated when the reaction takes place (Rigden, 2008). It is known that HAP domain can be found in multiple inositol polyphosphate phosphatase (MIPP) that responsible to dephosphorylate the higher species inositol phosphate $IP_6$ to $IP_3$ through $IP_5$ and $IP_4$ intermediates (Craxton *et al.*, 1997; Harwood, 2011). The fact that high species of inositol phosphates are elevated in the presence of lithium is in parallel with the suppression of protein possessing HAP domain that works on the dephosphorylation of high inositol phosphates to low species of inositol phosphates. This evidence implies beside inositol monophosphatase repression, lithium action can affect broader spectrum of inositol phosphate including attenuation of MIPP.

In *S. commune*, lithium does not only affect the growth, but also the sexual development. particularly the fruiting body development. Mating is not affected by the presence of lithium, since clamp connection could be observed under the microscope. The fruiting body is normally produced after 10-14 days post inoculation, but this phenomenon does not occur in the lithium treated mating plate. This results clearly indicates there must be an unknown mechanism by lithium that suppressing fruiting bodies development. The colony of mating plate treated by lithium shows irregular colony, some section of the colony produces flat mycelia and some section shows the normal fluffy mycelia making it irregular colony. The mating plate treated with lithium shows a melanin-like or a dark brown pigmentation indicating stress. It seems that the mating plate shows a stress response under lithium presence. Melanin is involved in the ability of fungi to survive in severe environments and providing protective role for fungi to adapt in the harsh condition (Eisenman and Casadevall, 2012). Melanin is a stresses scavenger and protects cell viability. Melanin was presumed to work as a shield against hydrolytic enzymes that may affect fungal cell walls integrity (Toledo *et al.*, 2017), which is in line with the proteomic result showing the repression of candidate chitin-binding protein in the presence of lithium. Similar finding showed also in another study by Mateus and colleagues (2014). Modification of colony color suggests that some fungi produced certain compounds in order to grow in the presence of lithium (Mateus *et al.*, 2014). However, the cause of pigmentation on the plate has to be investigated in the next future. Other stress-induced proteins that are found repressed as well under lithium treatment, are mitochondrial aconitase and aldo-keto reductase. Aldo-keto reductase protein catalyzes redox transformations and it is involved in biosynthesis, intermediary metabolism, and detoxification. Further, mitochondrial aconitase is involved in carbohydrate metabolism, catalyzing the second step of the tricarboxylic acid (TCA) cycle to generate isocitrate from the citrate precursor. Beside its role in energy production from the breakdown of carbohydrates, several intermediates of the TCA cycle are also biosynthetic precursors of haem and amino acids. Moreover, aconitase has another secondary role in the sustainability of mitochondrial DNA. On the other hand, it is hypothesized in *S. cerevisiae* that aldo-keto reductase might contribute to survival under stress circumstances (Ford and Ellis, 2001).

In parallel with this study, similar effect was found also in *S. cerevisiae*. Growth and progression through the cell cycle were affected by lithium and lithium caused yeast cells to arrest in G1 phase and affects negatively a-factor pheromone, suggesting that lithium alters the mating response and cell cycle of *S. cerevisiae* (Smith *et al.*, 1995). Lithium has influences on several other organisms and it may have various physiological targets. Development on slime mold *Dictyostelium,* Zebrafish, *Xenopus,* and sea urchin were affected by therapeutic relevant concentration of lithium, causing a teratogenic effect (Berridge *et al.*, 1989). In conclusion, lithium inhibits the growth and development in basidiomycetes, other fungi, and eukaryotes by affecting several carbohydrate & amino acid metabolisms and cellular signaling processes.

## 4.3 The role of Inositol monophosphatase

### 4.3.1 Inositol monophosphatase and inositol generation

Inositol monophosphatase is an enzyme that is responsible for the generation of the simple sugar polyol called inositol from inositol monophosphate. Inositol monophosphatase recycles the water-soluble phosphatidylinositol-phospholipase C (PI-PLC) degradation products, inositol triphosphates, to inositol, thus this enzyme maintains a moderate level of inositol pool.

In fact, there are two main sources by which fungal cells can obtain inositol. First, is to synthesize inositol internally in the cell. Intracellular glucose can be converted into inositol, which is the most common biologically active inositol isomer in *de novo* inositol biosynthetic pathway. Myo-inositol 1-phosphate synthase is the key enzyme in the conversion of glucose 6-phosphate to inositol monophosphate, subsequently the next step is performed by inositol monophosphatase, which dephosphorylates inositol 1-phosphate to *myo*-inositol. Inositol might as well be imported from the extracellular environment using inositol transporters (ITRs)(Xue, 2015). Inositol is an essential nutrient that is used for generating either the water soluble inositol phosphates or lipid inositol phosphates termed as phosphatidylinositol and its derivatives in eukaryotes (Reynolds, 2009). In *S. commune,* there are nine putative proteins which are similar with the protein that encodes inositol monophosphatase gene (*imp*), nevertheless these nine putative proteins do not possess high similarity in the three unique motifs so-called motif A, motif B, and motif C, therefore the gene that encodes these nine proteins are classified in the inositol monophosphatase gene like family. However, further investigation must be demonstrated to proof if these nine candidates are not responsible for the inositol generation. In the yeast *S. cerevisiae*, inositol monophosphatase plays important role in yeast growth, stress tolerance or inositol prototrophy, and indirectly activating the calcium signaling by Ena1 cation-extrusion pump (Lopez *et al.*, 1999). To gain a comprehensive knowledge of inositol monophosphatase, the attenuation of inositol monophosphatase by lithium and overexpression of inositol monophosphatase was performed. The inactivation of inositol monophosphatase by lithium, reduces enzyme activity significantly. Lithium interacts with a post-catalytic complex and occupies the binding site of the enzyme. which is supposed to be occupied by magnesium ion. The reduction of inositol monophosphatase activity is concomitant with reduction of *S. commune* growth, not only growth reduction which seems to be the effect of lithium to the fungi, but also, the retarded fungal growth shown by the lithium-treated fungi. Insignificant reduction of inositol was observed during the lithium treatment. There might be three reasons that is why inositol level was not impressively reduced. First, the technical problem during the inositol and inositol phosphate extraction. Mycelia grown in the liquid media grew floated in the liquid media and after pelleting, some of mycelia remained floated in the liquid media, therefore it was hard to wash the sample properly before inositol and inositol phosphates were extracted. Second reason is inositol perhaps can also be transported from the extracellular environment using specific inositol transporters (ITRs) or any sugar transporter. Third possible reason is, it might be that the incubation time for lithium in fungal cultivation was not sufficient, thus the inositol level was not significantly declined in the lithium treated fungi.

Inositol monophosphatase has high similarity to the well identified bovine inositol monophosphatase (Btimp) and *S. cerevisiae* inositol monophosphatase (Scimp). Majority of the identical or similar amino acid residues are at the active sites marked as motif A, B, and C, as it was determined from the crystal structure of the bovine enzyme. These residues have a similar functional role in the structure and catalytic mechanism of the methanogen enzymes (Chen and

Roberts, 1998). The active site of the bovine enzyme based on the X-ray crystallography data, includes the inositol binding site and two catalytic metal binding sites. According to the bovine inositol monophosphatase crystal structure, there are several amino acid residues that are important for binding capability, that could be found in *S. commune* inositol monophosphatase (Schimp) (see Table 12). The similarity of three unique motifs can be observed in plants, bacteria, and human as well.

### 4.3.2 Inositol monophosphatase is important for intracellular trafficking

To explore the functional role of inositol monophosphatase in *S. commune*, therefore inositol monophosphatase was overexpressed in this particular fungus. There is no difference in morphology of inositol monophosphatase overexpressed mutants with the control and parental strain. The growth rate between all of them are also almost identic. However, in the proteome level, we could distinguish the difference between the overexpressing inositol monophosphatase mutant and the control strain. There are 43 cellular processes and signaling associated proteins induced in *imp* overexpressing mutant compared to the control strain. The most striking features are the membrane trafficking machinery associated proteins and the proteins that are associated with the cytoskeleton regulation. This fact indicates that, membrane trafficking seems to be controlled by the inositol based signaling pathway. Inositol monophosphatase overexpression leads to the rapid inositol phosphates elevation, except inositol pentakisphosphate a ($IP_{5a}$ and $IP_{5b}$ species which are known to be the dephosphorylated products from $IP_6$). Signal recognition particle, SRP 19 subunit and SRP72 subunit are induced in the *imp* overexpressing mutant. SRP subunits are multimeric proteins that play role in targeting secretory proteins to the rough endoplasmic reticulum (RER) membrane in eukaryotes (Reyes *et al.*, 2007; Römisch *et al.*, 2006). Localization of proteins to their cellular destinations is important to sustain the arrangement and organization in the cell. SRP requires GTP for its activation. In the presence of GTP, SRP receptor binding to SRP results in the dissociation from both the signal sequence and the ribosome. Subsequently, GTP is hydrolyzed in order for the SRP to be released from the SRP receptor and released to the cytosol (Miller *et al.*, 1993). The induction of SRP19 subunit is in line with the induction of GTP-binding protein, HSR1-related RNA recognition motif, RNP-1. Snf7 is a component of the ESCRT (endosomal sorting complex required for transport)-III complex that is needed for the sorting and concentration of certain proteins resulting in the entry of the proteins into the invaginating vesicles of the multivesicular body (MVB). The ESCRT complexes are released from the endosomal membrane by the help of AAA-ATPase for further rounds of membrane invaginating (Babst *et al.*, 1998). AAA+-type ATPase containing the peptidase M41 domain is also up-regulated in the inositol monophosphatase overexpressing mutant in coincidence with the induction of the Snf7. ABC transporter which is responsible in the export-import of a wide range of substrates including small ions and macromolecules, is also induced in the *imp* overexpressing mutant. Nuclear transport factor 2 (NTF2) facilitates the import of GDP-bound Ran from the cytoplasm into the nucleus. NTF2 plays a crucial role for the function of Ran in shipment the receptor-mediated nucleocytoplasmic transport. NTF2 interrelates with GDP-bound Ran in the cytoplasm and recruits Ran to the nuclear pore complex and stimulates the nuclear import (Steggerda *et al.*, 2000; van Impe *et al.*, 2008). Protein transport SEC7-like and t-SNARE are also up-regulated in *imp* overexpressing mutant. SEC-7 is involved in vesicular budding and intracellular trafficking between compartments of the Golgi apparatus, on the other hand target membrane bound SNARE (t-SNARE) plays an important role in facilitating the vesicle fusion. SNAREs organize the cargo trafficking between the various organelles of the endomembrane machinery, however the SNAREs are not able to act alone, several other factors are required to mediate their function (Sanderfoot and Raikhel, 1999). In line with the elevation of membrane trafficking machineries associated proteins, cytoskeleton motor related proteins are also induced. This fact is convincing, since the intracellular trafficking is dependent on the cytoskeleton motor. Vesicle budding, and its movement involves actin cables to be associated with actin-based proteins. Cytoskeleton motor proteins listed here such as dynamin along with actin-binding, cofilin/tropomyosin, kinesin-like protein, and two other predicted actin bundling proteins, are induced in *imp* overexpressing mutants. Actin cytoskeleton is needed for a variety of cellular processes related to the membrane dynamics, including endocytosis, exocytosis, and vesicular trafficking at the Golgi compartment. All these occasions are regulated by complex networks of associated proteins and several of them are functionally linked to cell migration. Actin polymerization is involved in vesicle budding and fusion and this event is tightly controlled by small GTPases and the dynamin (Eitzen, 2003; Lanzetti, 2007). Kinesin and dynein are motor proteins involved in the movement of membrane vesicles along microtubules to and from cellular membrane section (Hamm-Alvarez, 1998). Since it has been found that intracellular trafficking is associated with inositol monophosphatase, therefore the effect of Brefeldin A (BFA) to the *imp* overexpressing mutants was further examined. BFA is a strong inhibitor of protein secretion in eukaryotic cells including filamentous fungi. Nevertheless, there was no clear different of *imp* overexpressing mutants in comparison to the control strain regarding to BFA effect. Although typical large vacuoles which are more abundance in *imp* overexpressing mutants were observed compared to the control strains. It is in parallel with the proteome analysis that cellular trafficking machinery in the *imp* overexpressing mutants are higher in abundance.

### 4.3.3 Potential role of inositol monophosphatase in cell wall integrity (CWI)

The fungal cell wall is a highly dynamic structure that is important for protecting the cell from changes in extracellular osmotic condition and it is also crucial for cell enlargement during development and morphogenesis. According to KOG description on proteomics analysis, chitinase, beta-1,6-N-acetylglucosaminyltransferase, and predicted endo-1,3-beta-glucanase are induced in inositol monophosphatase overexpressing mutants. The cell wall is loosened during polarized hyphal growth by several digestive enzymes (such as glucanases and chitinases) and enlarged at the hyphal tips and on the entire cell surface. The cell wall remodeling is executed in a highly regulated manner. The site of cell wall growth is loosened, allowing the cell expansion but not leading to the cell damage (Levin, 2005). Organization of the motor actin cytoskeleton and secretory vesicle targeting to the growth site are required for cell wall enlargement and expansion. Cell wall integrity (CWI) signaling pathway consists of a family of cell surface sensors that associated to a small G-protein Rho1. Rho1 activates a set of effectors and these effectors modulate a multitude set of cellular processes such as β-glucan synthesis at the site of cell wall remodeling, expression of cell wall biogenesis related gene, actin organization, and vesicular trafficking. In agreement with the previous study, Ras-related small GTPase, Rho type, is also induced in the *imp* overexpressing strain, in concomitant with the induction of digestive enzymes that are required for the cell wall expansion. Activation of Rho is also dependent of phosphatidylinositol 4,5 bisphosphate (PI 4,5-$P_2$). The evidence indicates that overexpression of inositol monophosphatase which results on the inositol level elevation increase possibly the production of PI 4,5-$P_2$. Elevation of PI 4,5-$P_2$ affects CWI signaling including up-regulation of Ras-related small GTPase, Rho. Several cell wall and membrane degrading chemicals including Calcofluor white, Congo red, SDS, Fenchlorazole and Tebuconazole were tested in the previous study (Traxler, 2017) and in this study. It has been observed that *S. commune imp* overexpressing mutants and control strains were sensitive to all of these stressors in certain concentrations. The *imp* overexpressing strains are less susceptible to the 500 µg/ml Congo red and 20 µg/ml SDS in comparison to the control strains. However, Calcofluor white, Fenchlorazole, and Tebuconazole do not show significant effect among tested fungi. The overall evidences show that inositol monophosphatase overexpression affects the cell wall integrity signaling.

## 4.4 Ras signaling is interrelated with inositol phosphate signaling

### 4.4.1 Possible crosstalk between Ras and inositol signaling

In the previous microarray study, it has been found that inositol phosphate signaling proteins, inositol monophosphatase and phosphatidylinositol synthase, are repressed in a Ras1 active mutant (Knabe *et al.*, 2013). The putative interrelation between inositol phosphate and Ras1 was further examined. Inositol monophosphatase gene expression was analyzed and confirmed via RT-qPCR and the result showed astonishing down-regulation of inositol monophosphatase transcription level when Ras1 is constitutively active. The *imp* gene down-regulation was shown in the constitutive active Ras1 mutant and in the deletion of Rasgap1 mutant. Gap1 is the activating protein of Ras1, therefore the deletion of Gap1 will lead to the constitutive expression Ras1. In overexpression of *imp* mutants, the *ras1* gene expression is depleted significantly but not the *ras2* gene expression. Therefore, we could conclude that there must be an unknown putative cross-talk between Ras1 and Imp. Previous report shows that the inositol (1,4,5) trisphosphate 3-kinase (ITP3K) has the potential to control Ras status through numerous targets within the regulatory network. ITP3K activation potentially increases Ras-GTP via repression of calcium signals that works on the activation of the Calcium-sensitive Ras-GAPs or stimulate Ras inactivation through the repression of calcium signals that activate the Calcium-sensitive Ras-GEFs or by activation of the $Gap1^{IP4BP}$ (Stokes *et al.*, 2006). However, the calcium signal was not examined in this study. Another previous study suggests that inactive state of $Gap1^{IP4BP}$ might bind to phosphatidylinositol 4,5-bisphosphate ($PIP_2$) via its PH domain and therefore localized to the plasma membrane. Localization of $PIP_2$ increases inositol tetrakisphosphate ($IP_4$) resulting in binding of $IP_4$ to $GAP1^{IP4BP}$ and stimulation of the GAP activity (Cozier *et al.*, 2000a; Cozier *et al.*, 2000b; Cullen *et al.*, 1995). The fact that $IP_4$ activation by ITP3K is in parallel with Ras activation goes hand in hand with the repression of inositol monophosphatase. The Ras activation prefers the phosphorylation of inositol polyphosphates from $IP_3$ rather than dephosphorylation cascade of $IP_3$ to lower inositol phosphate species such as $IP_2$, $IP_1$, and inositol. This phenomenon explains why the inositol monophosphatase is repressed in the Ras1 active manner. On the other hand, another early study shows that Ras signaling can be interrelated with the phosphatidylinositol 3-kinase (PI3K). PI3K has been known as one of the main effector pathways of Ras that modulate cell growth, cell cycle entry, survival of the cell, cytoskeleton motor arrangement, and metabolism (Castellano and Downward, 2010; Castellano and Downward, 2011; Vivanco and Sawyers, 2002). The possible link of Ras1 and inositol phosphate in the sexual development of *S. commune* was examined. The fact that Ras1 suppress the inositol monophosphatase, might be compensated with the overexpression of inositol monophosphatase. The mating and fruiting body development experiment of Ras1 active mutants with the inositol monophosphatase overexpressing mutants was established. Unfortunately, the result of mating is hard to interpret since there was no clear difference in the dikaryotic phase and fruiting bodies production between control strains and the inositol monophosphatase overexpressing strains.

### 4.4.2 Lithium action in Ras and inositol phosphate signaling

As it has been mentioned, that lithium is used as an inhibitor of inositol monophosphatase. The effect of lithium in the Ras1 active manner was examined. Inositol phosphates profile is not altered after Ras1 active mutants was treated with lithium for 24 hours, even though the incubation of cell with lithium is extended to 48 hours. On the other hand, the entire inositol phosphates profile was altered in the wild type treated with lithium, in particular the high species of inositol phosphates and the high energy inositol pyrophosphates $IP_7$ and $IP_8$. Ras1 activation is concomitant with the dramatically suppression of the *imp* gene expression and precludes inositol monophosphatase inhibition by lithium. Lithium inhibition of inositol monophosphatase that leads to a dramatic increase on higher phosphorylated inositol phosphate species occurs only in the wild type. These observations suggest that the presence of cellular inositol monophosphatase may affect the macromolecular complex of inositol phosphate kinases. If this is the case, inositol monophosphatase blockage by lithium will channel its substrate $IP_1$ toward the kinases synthesizing higher phosphorylated form of inositol. The tight transcriptional control by Ras1 on *imp* gene is most likely results in the drastic reduction of inositol monophosphatase activity in Ras1 active mutant. The absence of inositol monophosphatase in Ras1 active mutant from the hypothesized macromolecular complex prevents the substrate to be phosphorylated by the inositol phosphate kinases and therefore the side effect is the hyper synthesis of high phosphorylated forms of inositol phosphates which was observed in the wild type after lithium treatment, but not in Ras1 active mutant.

## 4.5 Sexual development of *S. commune*

The first comparative proteome analysis of all developmental stages in *S. commune*, from the haploid monokaryotic mycelium, dikaryotic mycelium, early stage of fruiting body (primordia), and mature fruiting body of *S. commune*, was performed. Alterations of proteomics level in comparison to the parental strains at various developmental stages using a powerful tool LC-MS/MS was analyzed to find out which proteins are regulated, and which certain chemical substances produced at which developmental stage, thus obtained data will provide a better understanding of machineries involved in hyphal growth and fruiting body formation of *S. commune*. Basidiospores of *S. commune* produce a vegetative monokaryotic mycelia. Monokaryotic mycelia later becomes fertile upon fusion with a compatible mating type partner, resulting in the initiation of dikaryon. Dikaryotic mycelia shows a shift from regular concentric growth to irregular growth of mycelia. Thereafter, aerial hyphae produce hyphal aggregates that develop into primordia. Primordia further differentiate to mature fruiting bodies containing basidiospores.

In dikaryon, there are 43 proteins and 26 proteins which are higher in abundance compared to monokaryon 12-43 and monokaryon 4-39, respectively, and also there are 97 and 94 are lower in abundance compared to monokaryon 12-43 and 4-39. However, only seven proteins that are found similarly down-regulated in comparison to both monokaryon. Cytochrome c is induced in dikaryon in comparison to monokaryon, indicating that apoptosis pathway could be activated, since it has been known in other fungi *S. cerevisiae* that cytochrome c is apoptotic-inducing stimulus (Roucou *et al.*, 2000) and mammalian cytochrome c is involved in the formation of apoptosomes in ATP-dependent manner (Zhou *et al.*, 2015a). Programmed cell death (apoptosis) is a highly regulated physiological activity and also a critical part of development that is performed alongside mitosis and differentiation in tissue development. Apoptosis is evolutionally conserved cell mechanisms found in multicellular eukaryotes and it is important for developmental process that upholds tissue homeostasis in higher multicellular organisms, whereas old or damaged cells are replaced by new cells (Ameisen, 2002; Yuan, 2006). Biochemical and morphological changes take place after a cell is defined to die (Davis *et al.*, 1996), which is in agreement to our finding that cytochrome c, the apoptosis regulator, was induced in the dikaryon. This finding shows that a switch in sexual differentiation requires apoptosis before morphological change takes place. Sugar transporter is found to be induced in the dikaryon. As it has been known that sugar transporters are antiporters that plays a crucial role in the biosynthesis of glycoconjugates and polysaccharides. Sugar transporters are ubiquitous in eukaryotes and they are important for the transport of nucleotide sugars from the cytosolic part to the organelles, such as golgi apparatus and endoplasmic reticulum. Alteration in the glycosylation of glycoproteins and glycolipids as well as in polysaccharide structure, resulting in the alterations in development (Handford *et al.*, 2006; Liu *et al.*, 2010). Mutation of GDP-mannose transporters (GmtA) in *A. nidulans* shows defect in hyphal morphological and alters cell wall properties, indicating that sugar transporters, in particular GDP-mannose transporters might be involved in fungal morphogenesis (Jackson-Hayes *et al.*, 2008). Two type of peptidases, peptidase M28 and peptidase M36, were induced in dikaryon in comparison to monokaryon. Peptidases are probably required during the switch from monokaryon to dikaryon, as it was shown also in the slime mold *Dictyostellium discoideum* that leucine aminopeptidase activity increases during morphogenesis (Firtel and Brackenbruy, 1972). Polysaccharide deacetylase was found to be induced in dikaryon, as well as in primordia and fruiting bodies. Polysaccharide deacetylase has been known to play a pivotal role in the alteration of cell shape and remodeling, implicating that polysaccharide deacetylase is responsible in hyphal morphogenesis (Arnaouteli *et al.*, 2015; Young, 2006). Polysaccharide deacetylase is known as chitin deacetylase, encompassing polysaccharide deacetylase domain in N-terminal and chitin binding module (CBM) on C-terminal of the protein (Cord-Landwehr *et al.*, 2016). A long-chain polymer chitin consists of β-1,4-linked *N*-acetylglucosamine, which is distinctive amongst the other polysaccharide components of the fungal cell wall and it is able to be modified chemically *via* deacetylation. Chitin deacetylation is catalyzed by chitin deacetylase (CDA) which belongs to the carbohydrate Esterase 4 family. It has been proposed in previous study of Geoghegan and Gurr (2017) that chitin deacetylase might be important for the cell wall integrity, cell wall flexibility, and cellular morphogenesis. Multicopper oxidase (MCO) has been found also up-regulated in dikaryon stage, primordia and fruiting body. MCO is able to catalyze the oxidation of various aromatic substrates in nature. It is involved in biodegradation of biopolymers such as lignin and have been shown to play pivotal roles during life cycles of fungi, especially in fruiting body development and pigment formation (Pöggeler, 2011).

In dikaryon, the fusion of two compatible mating or so-called anastomoses, takes place. Anastomoses might occupy several machineries in degradation of cell wall proteins and in degradation of chitin as fungal cell wall components. It is in line with the proteome analysis result, in which it was found that several important enzymes for polysaccharide and protein degradation, are higher induced in dikaryon hyphae.

Despite of several proteins' induction, there are even higher number of repressed proteins in dikaryon. Two membrane-integral ATP Binding Cassette (ABC) transporters are repressed in dikaryon. ABC transporters is omnipresent in eukaryote and it has been predicted as an ancient protein machinery for soluble material to pass a lipid bilayer through a concentration gradient (Higgins, 1992). This finding is convincing since the cell wall protein is degraded during dikaryotic phase, therefore the protein transport via ABC transporter is not required, leading to the repression of ABC transporter. Furthermore, aminoacyl-tRNA synthetase (AARS) and ribosomal proteins (ribosomal L23 protein, ribosomal protein S16, and ribosomal protein S27e) are depleted in dikaryon. As it has been known, AARS is the enzyme that catalyzes the aminoacylation reaction by covalently bound an amino acid to its cognate tRNA in the initiation of protein translation (Rajendran *et al.*, 2018), whereas ribosomal proteins catalyze mRNA-directed protein synthesis and known as essential house-keeping roles in ribosome biogenesis (Zhou *et al.*, 2015b). Presumably, cell wall protein biosynthesis mechanism might be blocked since in dikaryon, cell wall proteins are degraded during dikaryotic phase. Several glycoside hydrolases, enzymes that hydrolyzes the glycosidic bond between two or more carbohydrates, or between a carbohydrate and a non-carbohydrate moiety, are repressed in dikaryon, as well as in primordia and fruiting bodies, indicating that glycoside hydrolase is required in the entire processes of morphogenesis. On the other hand, a large group of small extracellular protein lipocalin which has functions to modulate cell homoeostasis and immune response (Flower, 1996), as well as carrier proteins, are also repressed in dikaryon. Protein kinase, Ste7-like protein which is associated in MAP Kinase signaling is down-regulated in dikaryon. The fact that MAPK signaling is down-regulated in dikaryotic phase shows indirectly that MAPK is only required in the initiation of mating, in response to pheromone recognition (Raudaskoski and Kothe, 2010). Once mating takes place, the MAP kinase is deactivated.

Phosphatidylinositol-4-phosphate 5-kinase (PI4P5K) was repressed in dikaryotic phase compared to monokaryon 12-43. PI4P5K is a plasma membrane localized protein that has been known to be important in actin polymerization and play a role in morphogenesis of ascomycete *C. albicans*. PI4P5K is stimulated during the switch from yeast form to the hyphal form under certain circumstances (Hairfield *et al.*, 2002; Homma *et al.*, 1998). In primordia and fruiting bodies, cytochrome P450 protein was induced in comparison to monokaryon. The similar finding was shown also in the previous study from Pelkmans (2016) and it is presumed that cytochrome P450 enzymes has a role in fruiting body formation. The *eln2* gene encodes cytochrome P450 enzyme. The cytochrome P450 enzymes are required in the biological oxidative, peroxidative, and reductive metabolism of various compounds. The mutation of gene encodes cytochrome P450 in *C. cinerea*, affects cell morphogenesis and tissue organization in the primordia. Primordia possesses a short shaft, plump and bulgy alike morphology, resulting in the mature fruiting bodies that possesses short stipe. In the mutant, it showed only a small number of cells can differentiate into the stipe tissue (Muraguchi and Kamada, 2000). The induction of cytochrome P450 in primordia and fruiting bodies of *S. commune* indicates that cytochrome P450 is required during development of fruiting body.

Beside cytochrome P450, cytochrome b-245 is also induced in fruiting bodies. One of aldehyde dehydrogenase is also stimulated in the primordia and fruiting bodies. Aldehyde dehydrogenase that involved in glyoxylate pathway, is involved in indole metabolism and implicated in multicellular development which are crucial for fruiting bodies development in ascomycete *Stigmatella aurantiaca* and basidiomycete *Flammulina velutipes* (Stamm *et al.*, 2005; Yoon *et* *al.*, 2002). The main pathways of carbohydrate metabolism, namely glycolysis, is hypothesized to be involved in ectomycorrhiza and fruiting bodies development (Deveau *et al.*, 2008). The induction of aldehyde dehydrogenase is concomitant with the induction of membrane protein auxin efflux carrier in primordia and fruiting bodies. As it is known that auxin (indole-3 acetate) is produced by aldehyde dehydrogenase from indole-3 acetaldehyde. In fruiting bodies, only hexose catabolism takes place. Hexoses or their precursor and conversion products are transferred from the vegetative mycelium or monokaryon to the fruiting body, whereas the other sugars more likely supply energy for growth and maintenance of the vegetative mycelia (Patyshakuliyeva *et al.*, 2013). Chorismate synthase, an enzyme that catalyzes the latest step in the shikimate pathway resulting aromatic compounds such as the aromatic amino acids, was found also elevated in primordia and fruiting body, suggesting that this enzyme is required in the early of fruiting body development until fruiting body maturation.

The ubiquitous enzyme copper amine oxidase (CuAO) is induced only in the primordia stage. Prokaryotes have a well-defined functional role in the metabolism of primary amines as alternative of carbon and nitrogen source to support and maintain the growth. CuAO exhibits a wide diversity in substrate structure and physiological function, ranging from oxidation of histamine, involvement of cell signaling cascade in animals to secondary metabolism and programmed cell death in plants (Brazeau *et al.*, 2004). In addition, in primordia and in fruiting bodies, amine oxidase (not copper related amine oxidase) was found also to be up-regulated compared to monokaryon, showing that amine metabolic process is involved in the fruiting body development and maturation. In primordia phase compared to monokaryon 4-39, amino acid permease was induced, this protein is an integral membrane permease that involved in the transport of amino acids into the cell. Amino acid transport is important for the switch of mycelial phase to the fruiting body since there are some amino acid that are found more abundant in fruiting body compared to the mycelia. A previous study showed that the most essential amino acid was found in the basidiomycete mushrooms *Ganoderma lucidum* and *Volvariella volvacea* are phenylalanine, tryptophan, and valine, which is known as mushroom great nutrient source (Sornprasert, 1995).

Ras small GTPase and protein kinase are involved also in the primordia, implying Ras signaling is involved in the initiation of fruiting body formation. Compared to monokaryon 4-39, adenylate cyclase-associated CAP which is known to be involved in cAMP signaling, was found more abundance in primordia, consistent with the earlier study where cAMP and Ras signaling is activated in the fruiting body of ascomycete *Cordyceps sinensis* (Feng et al., 2017). The two major upstream signaling regulators of adenylyl cyclase, which are guanine nucleotide-binding protein (G-protein) beta subunit-like protein and GTP-binding protein Ran (Ras GTPase family protein) increased in proteome level of the fruiting body of *C. sinensis* (Feng *et al.*, 2017). In the recent study by Pelkmans (2016), it is shown that high carbon dioxide level is declined in all developmental stages of fruiting body development of *S. commune* and carbon dioxide is sensed via cAMP signaling. Elevation on cellular cAMP level takes place as a result of the activity of adenylate cyclase, which allows for the activation of cAMP-dependent protein kinase (PKA). Binding of cAMP to the regulatory subunits of PKA stimulates conformational changes leads to the release and activation of the catalytic subunits of PKA (Palmer and Horton, 2006). It is important to notice that regulator G-protein and fungal G-protein alpha subunit are down-regulated in the fruiting body, implying that the GPCRs is only activated only during fruiting body formation. Several glycoside hydrolases and enzymes that are crucial for degrading complex carbohydrate were also found to be repressed in primordia and fruiting bodies, which created a confusion how the mechanism of glycoside hydrolase works. Nutrient deprivation is possibly a primary stress in fruiting hyphae and catabolism of lipids accumulated in vegetative mycelium might sustain the constant carbon flux that is required for fruiting body development and maturation.

On the other hand, calcium signaling machineries such as calcium-binding EF-hand and cytoskeleton motor proteins such as tubulin and actin were repressed in primordia and in fruiting bodies, implying cytoskeleton motor proteins such as tubulin and actin-binding related proteins are not required in the primordia and fruiting body, but in monokaryotic hyphae, cytoskeleton motor proteins are crucial to establish hyphal polarity, for maintaining the hyphal tips, and to facilitate secretory vesicle transport to the tip. Moreover, kinesin motor region and myosin motor region head are repressed in fruiting body, showing that transport activity along the hyphae is minimalized in fruiting body. Repression of cytoskeleton related proteins in fruiting body is parallel to the repression of proteins associated with secretory machinery and membrane trafficking machinery such as clathrin, clathrin adaptor, clathrin propeller, dynamin, importin-beta, alpha/gamma adaptin, Vps16, and phox-like domain containing protein. Alteration in cell wall metabolism during fruiting body morphogenesis have been observed in this study, as well as for many fungal species (Wessels, 1994).

Inositol and phosphatidylinositol signaling were also supressed in the fruiting body, which is shown by the repression of myo-inositol-1-phosphate synthase, phosphatidylinositol 3- and 4-kinase, and protein that possessed histidine acid phosphatase domain. Myo-inositol-1-phosphate synthase is the enzyme that is responsible converting glucose 6-phosphate to inositol 1-phosphate. The inception of the phosphatidylinositol phosphates metabolism comes from the omnipresent phospholipid in eukaryotic cells present in multitude proportions according to the type of membrane. Phosphatidylinositol phosphates are all metabolized subsequently from phosphatidylinositol. Phosphatidylinositol 3- and 4-kinase is responsible for the synthesis of phosphatidylinositol 3-phosphate (PI-3-P), phosphatidylinositol 3, 4-bisphosphate (PI-3,4-P(2)), and phosphatidylinositol 3,4,5-trisphosphate (PI-3,4,5-P(3)). PI3 kinase is involved in multiple cellular functions, including cell growth control, modulation of cell cycle, and maintenance of telomere length (Castellano and Downward, 2010; García *et al.*, 2006; Méndez-Pertuz *et al.*, 2017).

Histidine acid phosphatase is a domain protein that is possessed by inositol polyphosphate kinase such as inositol phosphate 6-kinase. The inositol based signaling has been associated with the membrane trafficking (De Craene *et al.*, 2017; Saiardi *et al.*, 2002). Phosphoinositides are lipid molecules that play role in coordinating and regulating intracellular trafficking. The synthesis of multifarious phosphatidylinositol phosphates is temporally and spatially controlled by the interaction between phosphatidylinositol lipid kinases and phosphatidylinositol lipid phosphatases in response to different stimuli. Haploid mycelia possibly require various inositol signaling molecules to be activated, but not during the subsequent development such as dikaryotic mycelia, primordia, and fruiting bodies. Polarized growth and microtubule dynamics were also deactivated in concomitant with the down-regulation of inositol based signaling proteins and several proteins related to the membrane trafficking machinery.

There are 21 different ribosomal proteins that are repressed in fruiting bodies, indicating that synthesis of proteins is limited in fruiting bodies. Homeodomain protein, which was found active in clamp connection development and in primary pairing and migration of compatible nuclei, was repressed in fruiting bodies. Early study by Pelkmans (2016) shows that homeodomain Hom2 is important in maintaining the mycelium in the vegetative phase. Hom2 is regulated by carbon dioxide and this homeoprotein is a target for phosphorylation by Protein kinase A (PKA), therefore it is convincing that there is a repression of protein kinase associated with MAPK and Ras family proteins in fruiting bodies. It seems that various signaling such as cAMP, MAPK, Ras, and inositol-based signaling is only required in the vegetative mycelia or during sexual development, however they are not necessarily needed once fruiting body is fully developed.

# 5 Outlook

Inositol based signaling consists of two main cascades, the first one is the inositol phosphate lipid route and the second one is water soluble inositol phosphate route. The lipid route is often called as phosphoinositide signaling route, which is not the focused topic in the study.

This is the first study of water-soluble inositol phosphate signaling as a part of G-protein coupled receptors (GPCRs) signaling in *S. commune*. However, there are still a lot of unsolved questions regarding to the complexity of this signaling molecules.

The spatial and temporal remodeling of inositol phosphate is an intrinsic feature that makes the orchestration of inositol phosphates is important for multifaceted cellular functions. Inositol phosphate signaling is potentially linked to Ras signaling, however the mechanism is still unknown, therefore the future experiment to explore how the cross-talk occur between inositol phosphate signaling and Ras signaling is urged to perform. Inositol monophosphatase generates inositol from inositol monophosphate, therefore inositol can be used as a building block for lipid inositol phosphate (phosphoinositide), such as phosphatidylinositol, and with the help of kinases, phosphatidylinositol phosphate can be generated.

Phosphatidylinositol phosphates are associated with the membrane cell and have functional role in multifarious cellular processes. However, the role of sequential phosphorylated phosphatidylinositol phosphates (PIP) in basidiomycete *S. commune* are not determined yet. It will be a great idea to study the role of PIP in this fungus and the functional role of phospholipase C (PLC) that cleaves phosphatidylinositol 4, 5-bisphosphates (PI 4,5-$P_2$) into the prominent molecules inositol trisphosphate ($IP_3$) and diacylglycerol (DAG). The development of experimental approaches to detect the intrinsic dynamics of PIP will be a key to unravel the regulatory role of PIP. Additionally, the interrelation between G-alpha proteins and PLC regarding PI 4,5-$P_2$ cleavage should be proven experimentally using the protein-protein interaction method.

In addition to that, interaction between Ras signaling and phosphatidylinositol signaling through the Ras effector phosphatidylinositol 3 kinase (PI3K) is required to examine in *S. commune*. *In silico* experiment could also be a great idea to search putative binding of Ras to the PI3K and to analyze the catalytic and regulatory domain of PI3K. Important to notice that *S. commune* possesses two Ras protein, Ras1 and Ras2. The involvement of Ras in inositol phosphate signaling was only explored in Ras1, therefore it might be also a crucial point to perform a mutagenesis experiment of Ras2 and to find out whether Ras2 is also involved in inositol phosphate signaling.

In this study, it is found that high species of inositol polyphosphate is reduced in fruiting body development, suggesting there is involvement of inositol phosphate kinases and phosphatases regulation in fruiting body development. The role of inositol hexakisphosphate ($IP_6$) and the energy-rich signaling molecules inositol pyrophosphates ($IP_7$ and $IP_8$) as well as enzymes that are involved in their phosphorylation and dephosphorylation, must be explored in the future. It is suggested to perform gene knock out, gene knock down and/or mutagenesis experiment to study the role of other inositol phosphate phosphatases and kinases.

Moreover, the quick analysis in inositol phosphate detection using Polyacrylamide Gel Electrophoresis (PAGE) has to be optimized for *S. commune*. Inositol phosphate signaling perhaps can be interrelated with other signaling cascade such as cyclic AMP (cAMP) signaling. $IP_6$ has been hypothesized to act on proline rich region of proteins which are crucial for protein-protein interactions, and thus consecutively important to stimulate cAMP production and cell signaling to complete the developmental cycle.

The involvement of inositol phosphate in cellular trafficking through overexpression of inositol monophosphatase was performed in this study, however the exact regulatory role of inositol phosphate clustering in cellular trafficking must be established and it will absolutely open up an exciting perspective in the field. Inositol phosphate might play a role in organizing the different steps in carrier biogenesis.

Proteomic analysis showed that there is evidence of inositol monophosphatase plays significant role in cellular trafficking and microscopic analysis using Synapto-Red FM 4-64 shows that *imp* overexpressing mutants possesses mostly larger vacuoles in comparison to the control strains. However, further analysis to characterize the nature of the membranous structures using an electron microscopy is suggested to perform.

# 6 Summary

This is the first study of inositol based signaling in the basidiomycete *S. commune*. This study contributes to the current understanding of inositol based signaling and role of inositol monophosphatase. *In silico* analysis in conjunction with experimental assays, has proved vital information for the future description of function in the inositol based signaling.

The putative crosstalk between inositol monophosphatase and Ras1 has been confirmed during this study using RT-qPCR, leading to the conclusion that inositol phosphate signaling and Ras signaling are somehow linked, yet the mechanism is still unknown. For the first time, inositol phosphate in *S. commune* was extracted and analyzed via Strong Acid Exchange-High Liquid Performance Chromatography (SAX-HPLC). From the HPLC elution profile, it was observed that *S. commune* could metabolize inositol monophosphate ($IP_1$), inositol bisphosphate ($IP_2$), inositol trisphosphate ($IP_3$), inositol tetrakisphosphate ($IP_4$), inositol pentakisphosphate ($IP_5$), inositol hexakisphosphate ($IP_6$), diphosphoinositol pentakisphosphate ($IP_7$) and bis-diphosphoinositol tetrakisphosphate ($IP_8$).

Lithium, the inhibitor of inositol monophosphatase, alters the entire inositol phosphates profile and induces the production of higher inositol phosphate species including $IP_3$, $IP_4$, $IP_5$, $IP_6$ and the energy-rich signaling molecules inositol pyrophosphates ($IP_7$ and $IP_8$). The induction of high species inositol phosphates by lithium suggests the molecular complex of inositol phosphate kinases. Interestingly, lithium does not alter the inositol phosphates profile in the Ras1 active mutant, indicating that activation of Ras1 might block the inhibitory effect of lithium on inositol monophosphatase and thus, not allowing the hyper-phosphorylation of high species inositol phosphates. Lithium significantly reduces the fungal growth in the wild type is in line with the change of inositol phosphate profile, whereas this phenomenon does not occur in the active Ras1 mutant. On the other hand, lithium inhibits the fruiting body production and development of *S. commune*, but it does not inhibit the mating processes and clamp formation development. Inositol phosphate metabolism plays crucial role in the fungal development. Inositol phosphates are synthesized by the spatiotemporal regulated activity of phosphatases and kinases metabolizing enzymes. Mycelial development in form of monokaryon and dikaryon requires $IP_6$ and the inositol pyrophosphates ($IP_7$ and $IP_8$), whereas $IP_6$, $IP_7$, and $IP_8$ are reduced in the fruiting body.

Proteome analysis revealed important information about the regulation of inositol based signaling molecules. Histidine acid phosphatase domain that is possessed by inositol polyphosphates phosphatases, the enzyme that dephosphorylates inositol phosphate, was found in a lower abundance in fruiting bodies. In addition to that, lithium significantly reduces the fungal growth in the wild type, which is in line with the change of inositol phosphate profile, whereas this phenomenon does not occur in the active Ras1 mutant. Myo-inositol 1-phosphate synthase and phosphatidylinositol 3- and 4- kinase are also decreased in fruiting bodies. The reduction of inositol phosphate and phosphatidylinositol signaling proteins, shows that these two signaling cascades are suppressed when the fruiting body is fully completed.

On the other hand, overexpression of inositol monophosphatase in *S. commune* shows that inositol monophosphatase is important in cellular trafficking regulation. Proteomic analysis revealed the induction of several proteins that are involved in cellular trafficking together with the cytoskeleton motor proteins that support the transport and cargo of the vesicular and cellular trafficking along the hyphae. Hence, inositol monophosphatase can be associated with the hyphal growth and polarization. Under laser scanning microscopy, the morphology of membranous organelles in *imp* overexpressing mutants and controls are observed and large vacuoles which are more abundant in *imp* overexpressing mutants were shown.

# 7 Zusammenfassung

Bei der vorliegenden Dissertation handelt es sich um die erste Forschungsarbeit zur inositolbasierten Signaltransduktion im Basidiomyzeten *S. commune*. Des Weiteren befasst sich die Arbeit mit der Rolle der Inositolmonophosphatase bei der Signaltransduktion. Durch die Verbindung von *In-silico*-Analyse mit experimentellen Laboruntersuchungen konnten wichtige Erkenntnisse zur zukünftigen Beschreibung der inositolbasierten Signaltransduktion gewonnen werden.

Die mutmaßliche Wechselwirkung zwischen der Inositolmonophosphatase und Ras1 konnte im Rahmen dieser Forschungsarbeit mittels RT-qPCR bestetätigt werden. Dies suggeriert, dass eine Verbindung zwischen dem Inositolmonophosphatase und Ras1 existiert, wobei die genauen Mechanismen noch unbekannt sind. Im Rahmen dieser Arbeit wurde erstmalig die Inositolmonophosphatase aus *S. commune* extrahiert und mittels Strong Acid Exchange-High Liquid Performance Chromatography (SAX-HPLC) analysiert. Das Elutionsprofil zeigte, dass *S. commune* Inositolmonophosphatase ($IP_1$), Inositoldiphosphat ($IP_2$), Inositoltriphosphat ($IP_3$), Inositoltetrakisphosphat ($IP_4$), Inositolpentakisphosphat ($IP_5$), Inositolhexakisphosphat ($IP_6$), Diphosphoinositol-Pentakisphosphat ($IP_7$) und Bis-Diphosphoinositol-Tetrakisphosphat ($IP_8$) verstoffwechseln kann.

Lithium, ein Inhibitor der Inositolmonophosphatase, verursachte eine Änderung des gesamten Inositolphosphatprofils und induzierte die Produktion „höherer“ Inositolphosphatspezies wie $IP_3$, $IP_4$, $IP_5$, $IP_6$ und die energiereichen Inositolpyrophosphat-Signalmoleküle ($IP_7$ und $IP_8$). Die Induktion von hohen Inositphosphaten durch Lithium zeigt einen molekularen Komplex von Inositolphosphatkinasen an. Lithium veränderte allerdings nicht das Inositolphosphatprofil der aktiven Ras1-Mutante, was darauf schließen lässt, dass die Aktivierung von Ras1 ggf. die inhibitorische Wirkung von Lithium auf die Inositolmonophosphatase aufheben kann, und somit der Hyperphosphorylierung hoher Inositolphosphatspezies entgegenwirkt. Lithium führte zu einer signifikaten Verringerung auf das Pilzwachstum im Wildtyp, was auf das veränderte Inositolphosphatprofil zurückgeführt werden kann. Dieses Phänomen wurde jedoch nicht in der Ras1-Mutante beobachtet. Des Weiteren inhibierte Lithium die Entwicklung und Bildung von Fruchtkörpern bei *S. commune*; es gab jedoch keine Auswirkung auf den Kreuzungsvorgang und die Schnallenbildung. Die Verstoffwechselung von Inositolphosphaten ist von großer Bedeutung für die Entwicklung des Pilzes. Die Synthese von Inositolphosphaten wird durch die regulierte Aktivität metabolisierender Enzyme wie Phosphatasen und Kinasen kontrolliert. Für die Entwicklung des von monokaryotischen und dikaryotischen werden $IP_6$ und die Inositolpyrophosphate $IP_7$ und $IP_8$ benötigt, wohingegen die $IP_6$, $IP_7$, und $IP_8$ im Fruchtkörper reduziert sind.

Die Proteom-Analyse zeigte wichtige Informationen über die Regulation von Inositol-basierten Signalmolekülen. Die Histidinsäurephosphatase- Domäne der Inositolpolyphosphat-Phosphatasen, das Enzym, welches Inositphosphat dephosphoryliert, wurde in einer geringeren Häufigkeit im Fruchtkörper gefunden, so wie auch die Myoinositol-1-Phosphat-Synthase und die Phosphatidylinositol-3- und -4-Kinase. Die Verringerung der Inositolphosphat- und Phosphatidylinositol-Signalproteine zeigt, dass beide Signalkaskaden nicht aktiviert sind, wenn der Fruchtkörper ausgereift ist.

Andererseits zeigt sich, dass die Überexpression der Inositolmonophosphatase in *S. commune* eine wichtige Rolle in der Regulierung von Transportprozessen innerhalb der Zelle spielt. Proteom-Analysen zeigten die Induktion mehrerer Proteine, die am Zelltransport beteiligt sind, sowie die Induktion motorischer Zytoskelettproteine, die am Transport von Vesikeln und Vakuolen entlang der Hyphe mitwirken. Dementsprechend kann die Inositolmonophosphatase mit dem Wachstum und der Polarisierung der Pilzhyphe assoziiert werden. Mittels Laserscanning-Mikroskopie wurde die Morphologie von membranhaltigen Organellen in Mutanten mit *imp*-Überexpression im Vergleich zu einer Kontrollen beobachtet und es konnte gezeigt werden, dass große Vakuolen in *imp*-Überexpressionsmutanten häufiger vorhanden sind.

# 8 References

Ameisen, J.C., 2002. On the origin, evolution, and nature of programmed cell death: a timeline of four billion years. *Cell death and differentiation* 9, 367-393.

Arnaouteli, S., Giastas, P., Andreou, A., Tzanodaskalaki, M., Aldridge, C., Tzartos, S.J., Vollmer, W., Eliopoulos, E., Bouriotis, V., 2015. Two Putative Polysaccharide Deacetylases Are Required for Osmotic Stability and Cell Shape Maintenance in *Bacillus anthracis*. *The Journal of Biological Chemistry* 290, 13465-13478.

Ásgeirsdóttir, S.A., Schuren, F.H.J., Wessels, J.G.H., 1994. Assignment of genes to pulse-field separated chromosomes of *Schizophyllum commune*. *Mycological Research* 98, 689-693.

Asgher, M., Wahab, A., Bilal, M., Nasir Iqbal, H.M., 2016. Lignocellulose degradation and production of lignin modifying enzymes by *Schizophyllum commune* IBL-06 in solid-state fermentation. *Biocatalysis and Agricultural Biotechnology* 6, 195-201.

Atack, J.R., 1995. Inositol monophosphatase inhibitors: a novel treatment for bipolar disorder? *Biol Psychiatry* 37, 761-763.

Atack, J.R., Broughton, H.B., Pollack, S.J., 1995. Structure and mechanism of inositol monophosphatase. *FEBS letters* 361, 1-7.

Azevedo, C., Saiardi, A., 2006. Extraction and analysis of soluble inositol polyphosphates from yeast. *Nat Protoc* 1, 2416-2422.

Babst, M., Wendland, B., Estepa, E.J., Emr, S.D., 1998. The Vps4p AAA ATPase regulates membrane association of a Vps protein complex required for normal endosome function. *Embo j* 17, 2982-2993.

Bardwell, L., 2005. A walk-through of the yeast mating pheromone response pathway. *Peptides* 26, 339-350.

Baykov, A.A., Evtushenko, O.A., Avaeva, S.M., 1988. A malachite green procedure for orthophosphate determination and its use in alkaline phosphatase-based enzyme immunoassay. *Anal Biochem* 171, 266-270.

Bellettini, M.B., Fiorda, F.A., Maieves, H.A., Teixeira, G.L., Ávila, S., Hornung, P.S., Júnior, A.M., Ribani, R.H., 2016. Factors affecting mushroom *Pleurotus spp*. *Saudi Journal of Biological Sciences*.

Berggard, T., Szczepankiewicz, O., Thulin, E., Linse, S., 2002. Myo-inositol monophosphatase is an activated target of calbindin D28k. *J Biol Chem* 277, 41954-41959.

Berne, S., Križaj, I., Pohleven, F., Turk, T., Maček, P., Sepčić, K., 2002. Pleurotus and Agrocybe hemolysins, new proteins hypothetically involved in fungal fruiting. *Biochimica et Biophysica Acta (BBA) - General Subjects* 1570, 153-159.

Berridge, M.J., 1984. Inositol trisphosphate and diacylglycerol as second messengers. *Biochemical Journal* 220, 345-360.

Berridge, M.J., 1993. Inositol trisphosphate and calcium signalling. *Nature* 361, 315-325.

Berridge, M.J., 2009. Inositol trisphosphate and calcium signalling mechanisms. *Biochim Biophys Acta* 1793, 933-940.

Berridge, M.J., Downes, C.P., Hanley, M.R., 1989. Neural and developmental actions of lithium: a unifying hypothesis. *Cell* 59, 411-419.

Berridge, M.J., Irvine, R.F., 1989. Inositol phosphates and cell signalling. *Nature* 341, 197-205.

Bolker, M., 1998. Sex and crime: heterotrimeric G proteins in fungal mating and pathogenesis. *Fungal Genet Biol* 25, 143-156.

Bone, R., Springer, J.P., Atack, J.R., 1992. Structure of inositol monophosphatase, the putative target of lithium therapy. *Proc Natl Acad Sci U S A* 89, 10031-10035.

Bordier, C., 1981. Phase separation of integral membrane proteins in Triton X-114 solution. *J Biol Chem* 256, 1604-1607.

Boulianne, R.P., Liu, Y., Aebi, M., Lu, B.C., Kues, U., 2000. Fruiting body development in *Coprinus cinereus*: regulated expression of two galectins secreted by a non-classical pathway. *Microbiology (Reading, England)* 146 ( Pt 8), 1841-1853.

Bouzarelou, D., Billini, M., Roumelioti, K., Sophianopoulou, V., 2008. EglD, a putative endoglucanase, with an expansin like domain is localized in the conidial cell wall of *Aspergillus nidulans*. *Fungal Genet Biol* 45, 839-850.

Bradford, M.M., 1976. A rapid and sensitive method for the quantitation of microgram quantities of protein utilizing the principle of protein-dye binding. *Anal Biochem* 72, 248-254.

Brazeau, B.J., Johnson, B.J., Wilmot, C.M., 2004. Copper-containing amine oxidases. Biogenesis and catalysis; a structural perspective. *Archives of Biochemistry and Biophysics* 428, 22-31.

Bro, C., Regenberg, B., Lagniel, G., Labarre, J., Montero-Lomeli, M., Nielsen, J., 2003. Transcriptional, proteomic, and metabolic responses to lithium in galactose-grown yeast cells. *J Biol Chem* 278, 32141-32149.

Brown, K.M., Tracy, D.K., 2013. Lithium: the pharmacodynamic actions of the amazing ion. *Therapeutic Advances in Psychopharmacology* 3, 163-176.

Bustin, S.A., Benes, V., Garson, J.A., Hellemans, J., Huggett, J., Kubista, M., Mueller, R., Nolan, T., Pfaffl, M.W., Shipley, G.L., Vandesompele, J., Wittwer, C.T., 2009. The MIQE guidelines: minimum information for publication of quantitative real-time PCR experiments. *Clinical chemistry* 55, 611-622.

Castellano, E., Downward, J., 2010. Role of RAS in the regulation of PI 3-kinase. *Curr Top Microbiol Immunol* 346, 143-169.

Castellano, E., Downward, J., 2011. RAS Interaction with PI3K: More Than Just Another Effector Pathway. *Genes & Cancer* 2, 261-274.

Cenis, J.L., 1992. Rapid extraction of fungal DNA for PCR amplification. *Nucleic Acids Research* 20, 2380.

Cestari, I., Anupama, A., Stuart, K., 2018. Inositol polyphosphate multikinase regulation of Trypanosoma brucei life stage development. *Mol Biol Cell* 29, 1137-1152.

Chen, L., Roberts, M.F., 1998. Cloning and expression of the inositol monophosphatase gene from *Methanococcus jannaschii* and characterization of the enzyme. *Appl Environ Microbiol* 64, 2609-2615.

Colombo, R., Milzani, A., Donne, I.D., 1991. Lithium increases actin polymerization rates by enhancing the nucleation step. *Journal of molecular biology* 217, 401-404.

Colot, H.V., Park, G., Turner, G.E., Ringelberg, C., Crew, C.M., Litvinkova, L., Weiss, R.L., Borkovich, K.A., Dunlap, J.C., 2006. A high-throughput gene knockout procedure for *Neurospora* reveals functions for multiple transcription factors. *Proceedings of the National Academy of Sciences* 103, 10352-10357.

Cord-Landwehr, S., Melcher, R.L.J., Kolkenbrock, S., Moerschbacher, B.M., 2016. A chitin deacetylase from the endophytic fungus *Pestalotiopsis sp.* efficiently inactivates the elicitor activity of chitin oligomers in rice cells. *Scientific Reports* 6, 38018.

Cosgrove, D.J., 2005. Growth of the plant cell wall. *Nat Rev Mol Cell Biol* 6, 850-861.

Cozier, G., Sessions, R., Bottomley, J.R., Reynolds, J.S., Cullen, P.J., 2000a. Molecular modelling and site-directed mutagenesis of the inositol 1,3,4,5-tetrakisphosphate-binding pleckstrin homology domain from the Ras GTPase-activating protein GAP1IP4BP. *The Biochemical journal* 349, 333-342.

Cozier, G.E., Lockyer, P.J., Reynolds, J.S., Kupzig, S., Bottomley, J.R., Millard, T.H., Banting, G., Cullen, P.J., 2000b. GAP1IP4BP contains a novel group I pleckstrin homology domain that directs constitutive plasma membrane association. *J Biol Chem* 275, 28261-28268.

Craxton, A., Caffrey, J.J., Burkhart, W., Safrany, S.T., Shears, S.B., 1997. Molecular cloning and expression of a rat hepatic multiple inositol polyphosphate phosphatase. *Biochemical Journal* 328, 75-81.

Culbertson, M.R., Donahue, T.F., Henry, S.A., 1976. Control of inositol biosynthesis in *Saccharomyces cerevisiae*; inositol-phosphate synthetase mutants. *J Bacteriol* 126, 243-250.

Cullen, P.J., Hsuan, J.J., Truong, O., Letcher, A.J., Jackson, T.R., Dawson, A.P., Irvine, R.F., 1995. Identification of a specific Ins(1,3,4,5)P4-binding protein as a member of the GAP1 family. *Nature* 376, 527-530.

Culotta, V.C., 2001. Superoxide dismutase, oxidative stress, and cell metabolism, in: Stadtman, E.R., Chock, P.B. (Eds.), Current Topics in Cellular Regulation. Academic Press, pp. 117-132.

Dávila Costa, J.S., Albarracín, V.H., Abate, C.M., 2011. Responses of environmental *Amycolatopsis* strains to copper stress. *Ecotoxicology and Environmental Safety* 74, 2020-2028.

Davis, E.C., Popper, P., Gorski, R.A., 1996. The role of apoptosis in sexual differentiation of the rat sexually dimorphic nucleus of the preoptic area. *Brain Res* 734, 10-18.

De Craene, J.O., Bertazzi, D.L., Bar, S., Friant, S., 2017. Phosphoinositides, Major Actors in Membrane Trafficking and Lipid Signaling Pathways. *International journal of molecular sciences* 18.

de Jong, J.F., Deelstra, H.J., Wösten, H.A.B., Lugones, L.G., 2006. RNA-Mediated Gene Silencing in Monokaryons and Dikaryons of Schizophyllum commune. *Applied and Environmental Microbiology* 72, 1267-1269.

Deranieh, R.M., Shi, Y., Tarsio, M., Chen, Y., McCaffery, J.M., Kane, P.M., Greenberg, M.L., 2015. Perturbation of the Vacuolar ATPase: A novel consequence of inositol depletion. *The Journal of Biological Chemistry* 290, 27460-27472.

DeRisi, J.L., Iyer, V.R., Brown, P.O., 1997. Exploring the Metabolic and Genetic Control of Gene Expression on a Genomic Scale. *Science* 278, 680-686.

Deveau, A., Kohler, A., Frey-Klett, P., Martin, F., 2008. The major pathways of carbohydrate metabolism in the ectomycorrhizal basidiomycete *Laccaria bicolor* S238N. *New Phytol* 180, 379-390.

Di Paolo, G., De Camilli, P., 2006. Phosphoinositides in cell regulation and membrane dynamics. *Nature* 443, 651-657.

Dubois, E., Messenguy, F., 1994. Pleiotropic function of ArgRIIIp (Arg82p), one of the regulators of arginine metabolism in *Saccharomyces cerevisiae*. Role in expression of cell-type-specific genes. *Mol Gen Genet* 243, 315-324.

Dudev, T., Lim, C., 2011. Competition between Li+ and Mg2+ in metalloproteins. Implications for lithium therapy. *J Am Chem Soc* 133, 9506-9515.

Dutta, A., Bhattacharyya, S., Dutta, D., Das, A.K., 2014. Structural elucidation of the binding site and mode of inhibition of $Li^+$ and $Mg^{2+}$ in inositol monophosphatase. *The FEBS journal* 281, 5309-5324.

Efrati, S., Averbukh, M., Berman, S., Feldman, L., Dishy, V., Kachko, L., Weissgarten, J., Golik, A., Averbukh, Z., 2005. N-Acetylcysteine ameliorates lithium-induced renal failure in rats. *Nephrology, dialysis, transplantation : official publication of the European Dialysis and Transplant Association - European Renal Association* 20, 65-70.

Eger-Hummel, G., 1980. Blue-Light Photomorphogenesis in Mushrooms (Basidiomycetes), in: Senger, H. (Ed.), The Blue Light Syndrome. Springer Berlin Heidelberg, pp. 555-562.

Eisenman, H.C., Casadevall, A., 2012. Synthesis and assembly of fungal melanin. *Applied Microbiology and Biotechnology* 93, 931-940.

Eitzen, G., 2003. Actin remodeling to facilitate membrane fusion. *Biochimica et Biophysica Acta (BBA) - Molecular Cell Research* 1641, 175-181.

Erdmann, S., Freihorst, D., Raudaskoski, M., Schmidt-Heck, W., Jung, E.M., Senftleben, D., Kothe, E., 2012. Transcriptome and functional analysis of mating in the basidiomycete *Schizophyllum commune*. *Eukaryot Cell* 11, 571-589.

Feng, K., Wang, L.-y., Liao, D.-j., Lu, X.-p., Hu, D.-j., Liang, X., Zhao, J., Mo, Z.-y., Li, S.-p., 2017. Potential molecular mechanisms for fruiting body formation of *Cordyceps* illustrated in the case of *Cordyceps sinensis*. *Mycology* 8, 231-258.

Firtel, R.A., Brackenbruy, R.W., 1972. Partial characterization of several protein and amino acid metabolizing enzymes in the cellular slime mold *Dictyostelium discoideum*. *Dev Biol* 27, 307-321.

Flower, D.R., 1996. The lipocalin protein family: structure and function. *Biochemical Journal* 318, 1-14.

Ford, G., Ellis, E.M., 2001. Three aldo-keto reductases of the yeast *Saccharomyces cerevisiae*. *Chemico-biological interactions* 130-132, 685-698.

Freihorst, D., 2015. Microarray analyses and new pheromone receptors in *Schizophyllum commune*. Friedrich Schiller Universiät Jena.

Freihorst, D., Brunsch, M., Wirth, S., Krause, K., Kniemeyer, O., Linde, J., Kunert, M., Boland, W., Kothe, E., 2016. Smelling the difference: Transcriptome, proteome and volatilome changes after mating. *Fungal Genet Biol*.

Ganeshpurkar, A., Rai, G., Jain, A.P., 2010. Medicinal mushrooms: Towards a new horizon. *Pharmacognosy Reviews* 4, 127-135.

Gao, C., 2016. Application of a novel biopolymer to enhance oil recovery. *Journal of Petroleum Exploration and Production Technology* 6, 749-753.

García, Z., Kumar, A., Marqués, M., Cortés, I., Carrera, A.C., 2006. Phosphoinositide 3-kinase controls early and late events in mammalian cell division. *The EMBO Journal* 25, 655-661.

Geoghegan, I.A., Gurr, S.J., 2017. Investigating chitin deacetylation and chitosan hydrolysis during vegetative growth in *Magnaporthe oryzae*. *Cell Microbiol* 19.

Gietz, R.D., Schiestl, R.H., Willems, A.R., Woods, R.A., 1995. Studies on the transformation of intact yeast cells by the LiAc/SS-DNA/PEG procedure. *Yeast* 11, 355-360.

Gietz, R.D., Sugino, A., 1988. New yeast-Escherichia coli shuttle vectors constructed with in vitro mutagenized yeast genes lacking six-base pair restriction sites. *Gene* 74, 527-534.

Gola, S., Kothe, E., 2003. An expression system for the functional analysis of pheromone genes in the tetrapolar basidiomycete *Schizophyllum commune*. *Journal of basic microbiology* 43, 104-112.

Guse, A.H., Greiner, E., Emmrich, F., Brand, K., 1993. Mass changes of inositol 1,3,4,5,6-pentakisphosphate and inositol hexakisphosphate during cell cycle progression in rat thymocytes. *J Biol Chem* 268, 7129-7133.

Haimovich, A., Eliav, U., Goldbourt, A., 2012. Determination of the lithium binding site in inositol monophosphatase, the putative target for lithium therapy, by magic-angle-spinning solid-state NMR. *J Am Chem Soc* 134, 5647-5651.

Hairfield, M.L., Westwater, C., Dolan, J.W., 2002. Phosphatidylinositol-4-phosphate 5-kinase activity is stimulated during temperature-induced morphogenesis in *Candida albicans*. *Microbiology (Reading, England)* 148, 1737-1746.

Hamm-Alvarez, S.F., 1998. Molecular motors and their role in membrane traffic. *Advanced drug delivery reviews* 29, 229-242.

Hanakahi, L.A., Bartlet-Jones, M., Chappell, C., Pappin, D., West, S.C., 2000. Binding of Inositol Phosphate to DNA-PK and Stimulation of Double-Strand Break Repair. *Cell* 102, 721-729.

Handford, M., Rodriguez-Furlan, C., Orellana, A., 2006. Nucleotide-sugar transporters: structure, function and roles in vivo. *Brazilian journal of medical and biological research = Revista brasileira de pesquisas medicas e biologicas* 39, 1149-1158.

Harwood, A.J., 2005. Lithium and bipolar mood disorder: the inositol-depletion hypothesis revisited. *Molecular psychiatry* 10, 117-126.

Harwood, A.J., 2011. Prolyl Oligopeptidase, Inositol Phosphate Signalling and Lithium Sensitivity. *CNS & Neurological Disorders Drug Targets* 10, 333-339.

Higgins, C.F., 1992. ABC transporters: from microorganisms to man. *Annual review of cell biology* 8, 67-113.

Hirai, H., Kashima, Y., Hayashi, K., Sugiura, T., Yamagishi, K., Kawagishi, H., Nishida, T., 2008. Efficient expression of laccase gene from white-rot fungus *Schizophyllum commune* in a transgenic tobacco plant. *FEMS Microbiology Letters* 286, 130-135.

Homma, K., Terui, S., Minemura, M., Qadota, H., Anraku, Y., Kanaho, Y., Ohya, Y., 1998. Phosphatidylinositol-4-phosphate 5-kinase localized on the plasma membrane is essential for yeast cell morphogenesis. *J Biol Chem* 273, 15779-15786.

Høy, M., Efanov, A.M., Bertorello, A.M., Zaitsev, S.V., Olsen, H.L., Bokvist, K., Leibiger, B., Leibiger, I.B., Zwiller, J., Berggren, P.-O., Gromada, J., 2002. Inositol hexakisphosphate promotes dynamin I- mediated endocytosis. *Proceedings of the National Academy of Sciences* 99, 6773-6777.

Hsu, K.H., Lee, Y.R., Lin, Y.L., Chu, F.H., 2011. Cytochrome P450 genes in medicinal mushroom *Antrodia cinnamomea* T.T. Chang et W.N. Chou (higher Basidiomycetes) are strongly expressed during fruiting body formation. *International journal of medicinal mushrooms* 13, 513-523.

Hughes, A.R., Putney, J.W., Jr., 1990. Inositol phosphate formation and its relationship to calcium signaling. *Environ Health Perspect* 84, 141-147.

Irvine, R.F., 2016. A short history of inositol lipids. *Journal of lipid research* 57, 1987-1994.

Irvine, R.F., Schell, M.J., 2001. Back in the water: the return of the inositol phosphates. *Nat Rev Mol Cell Biol* 2, 327-338.

Jackson-Hayes, L., Hill, T.W., Loprete, D.M., Fay, L.M., Gordon, B.S., Nkashama, S.A., Patel, R.K., Sartain, C.V., 2008. Two GDP-mannose transporters contribute to hyphal form and cell wall integrity in *Aspergillus nidulans*. *Microbiology (Reading, England)* 154, 2037-2047.

Jung, E.M., Kothe, E., Raudaskoski, M., 2018. The making of a mushroom: Mitosis, nuclear migration and the actin network. *Fungal Genet Biol* 111, 85-91.

Kalujnaia, S., McVee, J., Kasciukovic, T., Stewart, A.J., Cramb, G., 2010. A role for inositol monophosphatase 1 (IMPA1) in salinity adaptation in the euryhaline eel (Anguilla anguilla). *FASEB J* 24, 3981-3991.

Kielczykowska, M., Pasternak, K., Musik, I., Wroniska, J., 2004. The effect of lithium administration in a diet on the chosen parameters of the antioxidant barrier in rats. *Annales Universitatis Mariae Curie-Sklodowska. Sectio D: Medicina* 59, 140-145.

Kirtzel, J., Scherwietes, E.L., Merten, D., Krause, K., Kothe, E., 2018. Metal release and sequestration from black slate mediated by a laccase of *Schizophyllum commune*. *Environmental Science and Pollution Research*.

Kiselyov, K., Shin, D.M., Muallem, S., 2003. Signalling specificity in GPCR-dependent $Ca^{2+}$ signalling. *Cellular Signalling* 15, 243-253.

Knabe, N., Jung, E.-M., Freihorst, D., Hennicke, F., Horton, J.S., Kothe, E., 2013. A Central Role for Ras1 in Morphogenesis of the Basidiomycete *Schizophyllum commune*. *Eukaryotic Cell* 12, 941-952.

Kobilka, B.K., 2007. G Protein Coupled Receptor Structure and Activation. *Biochimica et biophysica acta* 1768, 794-807.

Kothe, E., 1999. Mating types and pheromone recognition in the Homobasidiomycete schizophyllum commune. *Fungal Genet Biol* 27, 146-152.

Kothe, E., Erdmann, S., Knabe, N., Jung, E.M., 2009. Fungal GPCR signaling in pheromone response. *Cell Communication and Signaling : CCS* 7, A62-A62.

Kuo, H.F., Chang, T.Y., Chiang, S.F., Wang, W.D., Charng, Y.Y., Chiou, T.J., 2014. *Arabidopsis* inositol pentakisphosphate 2-kinase, AtIPK1, is required for growth and modulates phosphate homeostasis at the transcriptional level. *The Plant journal : for cell and molecular biology* 80, 503-515.

Lanzetti, L., 2007. Actin in membrane trafficking. *Curr Opin Cell Biol* 19, 453-458.

Larsson, O., Barker, C.J., Sjöholm, Å., Carlqvist, H., Michell, R.H., Bertorello, A., Nilsson, T., Honkanen, R.E., Mayr, G.W., Zwiller, J., Berggren, P.-O., 1997. Inhibition of Phosphatases and Increased Ca<sup>2+</sup> Channel Activity by Inositol Hexakisphosphate. *Science* 278, 471-474.

Lee, C.H., Dixon, J.F., Reichman, M., Moummi, C., Los, G., Hokin, L.E., 1992. Li+ increases accumulation of inositol 1,4,5-trisphosphate and inositol 1,3,4,5-tetrakisphosphate in cholinergically stimulated brain cortex slices in guinea pig, mouse and rat. The increases require inositol supplementation in mouse and rat but not in guinea pig. *The Biochemical journal* 282 ( Pt 2), 377-385.

Lepkifker, E., Iancu, I., Horesh, N., Strous, R.D., Kotler, M., 2007. Lithium therapy for unipolar and bipolar depression among the middle-aged and older adult patient subpopulation. *Depression and anxiety* 24, 571-576.

Lev, S., Li, C., Desmarini, D., Saiardi, A., Fewings, N.L., Schibeci, S.D., Sharma, R., Sorrell, T.C., Djordjevic, J.T., 2015. Fungal Inositol Pyrophosphate IP7 Is Crucial for Metabolic Adaptation to the Host Environment and Pathogenicity. *mBio* 6, e00531-00515.

Levin, D.E., 2005. Cell Wall Integrity Signaling in *Saccharomyces cerevisiae*. *Microbiology and Molecular Biology Reviews* 69, 262-291.

Li, L., Wright, S.J., Krystofova, S., Park, G., Borkovich, K.A., 2007. Heterotrimeric G protein signaling in filamentous fungi. *Annu Rev Microbiol* 61, 423-452.

Liu, L., Xu, Y.-X., Hirschberg, C.B., 2010. The role of nucleotide sugar transporters in development of eukaryotes. *Seminars in cell & developmental biology* 21, 600-608.

Lopez, F., Leube, M., Gil-Mascarell, R., Navarro-Aviñó, J.P., Serrano, R., 1999. The yeast inositol monophosphatase is a lithium- and sodium-sensitive enzyme encoded by a non-essential gene pair. *Molecular Microbiology* 31, 1255-1264.

Lu, S., Huang, W., Li, X., Huang, Z., Liu, X., Chen, Y., Shi, T., Zhang, J., 2012. Insights into the role of magnesium triad in myo-inositol monophosphatase: metal mechanism, substrate binding, and lithium therapy. *J Chem Inf Model* 52, 2398-2409.

Luo, H.R., Huang, Y.E., Chen, J.C., Saiardi, A., Iijima, M., Ye, K., Huang, Y., Nagata, E., Devreotes, P., Snyder, S.H., 2003. Inositol pyrophosphates mediate chemotaxis in *Dictyostelium* via pleckstrin homology domain-PtdIns(3,4,5)P3 interactions. *Cell* 114, 559-572.

Ma, Y., Lieber, M.R., 2002. Binding of Inositol Hexakisphosphate (IP6) to Ku but Not to DNA-PKcs. *Journal of Biological Chemistry* 277, 10756-10759.

Madhavan, S., Krause, K., Jung, E.-M., Kothe, E., 2014. Differential regulation of multi-copper oxidases in *Schizophyllum commune* during sexual development. *Mycological Progress* 13, 1009.

Majerus, P.W., Kisseleva, M.V., Norris, F.A., 1999. The role of phosphatases in inositol signaling reactions. *J Biol Chem* 274, 10669-10672.

Margelis, S., D'Souza, C., Small, A.J., Hynes, M.J., Adams, T.H., Davis, M.A., 2001. Role of Glutamine Synthetase in Nitrogen Metabolite Repression in *Aspergillus nidulans*. *Journal of Bacteriology* 183, 5826-5833.

Martins, L.F., Montero-Lomeli, M., Masuda, C.A., Fortes, F.S., Previato, J.O., Mendonca-Previato, L., 2008. Lithium-mediated suppression of morphogenesis and growth in Candida albicans. *FEMS Yeast Res* 8, 615-621.

Masuda, C.A., Ramirez, J., Pena, A., Montero-Lomeli, M., 2000. Regulation of monovalent ion homeostasis and pH by the Ser-Thr protein phosphatase SIT4 in *Saccharomyces cerevisiae*. *J Biol Chem* 275, 30957-30961.

Mateus, D.N., Willian, L.C., Jos, M.R.d.L., Maria, C.M.K., 2014. Lithium chloride affects mycelial growth of white rot fungi: Fungal screening for Li-enrichment. *African Journal of Microbiology Research* 8, 2111-2123.

Méndez-Pertuz, M., Martínez, P., Blanco-Aparicio, C., Gómez-Casero, E., Belen García, A., Martínez-Torrecuadrada, J., Palafox, M., Cortés, J., Serra, V., Pastor, J., Blasco, M.A., 2017. Modulation of telomere protection by the PI3K/AKT pathway. *Nature Communications* 8, 1278.

Menniti, F.S., Oliver, K.G., Putney, J.W., Jr., Shears, S.B., 1993. Inositol phosphates and cell signaling: new views of InsP5 and InsP6. *Trends Biochem Sci* 18, 53-56.

Miller, J.D., Wilhelm, H., Gierasch, L., Gilmore, R., Walter, P., 1993. GTP binding and hydrolysis by the signal recognition particle during initiation of protein translocation. *Nature* 366, 351-354.

Mullaney, E.J., Ullah, A.H.J., 2005. Conservation of cysteine residues in fungal histidine acid phytases. *Biochemical and biophysical research communications* 328, 404-408.

Munoz-Rivas, A., Specht, C.A., Drummond, B.J., Froeliger, E., Novotny, C.P., Ullrich, R.C., 1986. Transformation of the basidiomycete, *Schizophyllum commune*. *Mol Gen Genet* 205, 103-106.

Muraguchi, H., Kamada, T., 2000. A mutation in the eln2 gene encoding a cytochrome P450 of *Coprinus cinereus* affects mushroom morphogenesis. *Fungal Genet Biol* 29, 49-59.

Muraguchi, H., Umezawa, K., Niikura, M., Yoshida, M., Kozaki, T., Ishii, K., Sakai, K., Shimizu, M., Nakahori, K., Sakamoto, Y., Choi, C., Ngan, C.Y., Lindquist, E., Lipzen, A., Tritt, A., Haridas, S., Barry, K., Grigoriev, I.V., Pukkila, P.J., 2015. Strand-Specific RNA-Seq Analyses of Fruiting Body Development in *Coprinopsis cinerea*. *PLoS One* 10, e0141586.

Murray, M., Greenberg, M.L., 1997. Regulation of inositol monophosphatase in *Saccharomyces cerevisiae*. *Mol Microbiol* 25, 541-546.

Murray, M., Greenberg, M.L., 2000. Expression of yeast INM1 encoding inositol monophosphatase is regulated by inositol, carbon source and growth stage and is decreased by lithium and valproate. *Mol Microbiol* 36, 651-661.

Murray, M.G., Thompson, W.F., 1980. Rapid isolation of high molecular weight plant DNA. *Nucleic Acids Research* 8, 4321-4325.

Nayak, A.P., Green, B.J., Beezhold, D.H., 2013. Fungal hemolysins. *Medical mycology* 51, 1-16.

Neuwald, A.F., York, J.D., Majerus, P.W., 1991. Diverse proteins homologous to inositol monophosphatase. *FEBS letters* 294, 16-18.

Niculita-Hirzel, H.a.H., A., 2008. Visualizing the gene order conservation among genomes with ChromoMapper. *International Journal of Computational Intelligence in Bioinformatics and Systems Biology, 1.*

Nielsen, J., Hoffert, J.D., Knepper, M.A., Agre, P., Nielsen, S., Fenton, R.A., 2008. Proteomic analysis of lithium-induced nephrogenic diabetes insipidus: Mechanisms for aquaporin 2 down-regulation and cellular proliferation. *Proceedings of the National Academy of Sciences of the United States of America* 105, 3634-3639.

Norris, F.A., Ungewickell, E., Majerus, P.W., 1995. Inositol Hexakisphosphate Binds to Clathrin Assembly Protein 3 (AP-3/AP180) and Inhibits Clathrin Cage Assembly in Vitro. *Journal of Biological Chemistry* 270, 214-217.

Nourbakhsh, A., Collakova, E., Gillaspy, G.E., 2014. Characterization of the inositol monophosphatase gene family in *Arabidopsis*. *Front Plant Sci* 5, 725.

Ohm, R.A., Aerts, D., Wosten, H.A., Lugones, L.G., 2013. The blue light receptor complex WC-1/2 of Schizophyllum commune is involved in mushroom formation and protection against phototoxicity. *Environ Microbiol* 15, 943-955.

Ohm, R.A., de Jong, J.F., Berends, E., Wang, F., Wosten, H.A., Lugones, L.G., 2010a. An efficient gene deletion procedure for the mushroom-forming basidiomycete *Schizophyllum commune*. *World J Microbiol Biotechnol* 26, 1919-1923.

Ohm, R.A., de Jong, J.F., de Bekker, C., Wosten, H.A., Lugones, L.G., 2011. Transcription factor genes of *Schizophyllum commune* involved in regulation of mushroom formation. *Mol Microbiol* 81, 1433-1445.

Ohm, R.A., de Jong, J.F., Lugones, L.G., Aerts, A., Kothe, E., Stajich, J.E., de Vries, R.P., Record, E., Levasseur, A., Baker, S.E., Bartholomew, K.A., Coutinho, P.M., Erdmann, S., Fowler, T.J., Gathman, A.C., Lombard, V., Henrissat, B., Knabe, N., Kues, U., Lilly, W.W., Lindquist, E., Lucas, S., Magnuson, J.K., Piumi, F., Raudaskoski, M., Salamov, A., Schmutz, J., Schwarze, F.W., vanKuyk, P.A., Horton, J.S., Grigoriev, I.V., Wosten, H.A., 2010b. Genome sequence of the model mushroom *Schizophyllum commune*. *Nat Biotechnol* 28, 957-963.

Okazaki, M., Adachi, Y., Ohno, N., Yadomae, T., 1995. Structure-activity relationship of (1-->3)-beta-D-glucans in the induction of cytokine production from macrophages, in vitro. *Biological & pharmaceutical bulletin* 18, 1320-1327.

Palmer, G.E., Horton, J.S., 2006. Mushrooms by magic: making connections between signal transduction and fruiting body development in the basidiomycete fungus *Schizophyllum commune*. *FEMS Microbiol Lett* 262, 1-8.

Patyshakuliyeva, A., Jurak, E., Kohler, A., Baker, A., Battaglia, E., de Bruijn, W., Burton, K.S., Challen, M.P., Coutinho, P.M., Eastwood, D.C., Gruben, B.S., Makela, M.R., Martin, F., Nadal, M., van den Brink, J., Wiebenga, A., Zhou, M., Henrissat, B., Kabel, M., Gruppen, H., de Vries, R.P., 2013. Carbohydrate utilization and metabolism is highly differentiated in *Agaricus bisporus*. *BMC genomics* 14, 663.

Pelkmans, J.F., 2016. Environmental signaling and regulation of mushroom formation. Utrecht University.

Perkins, J.H., 1969. Morphogenesis in *Schizophyllum commune*. I. Effects of white light. *Plant Physiol* 44, 1706-1711.

Pisani, F., Livermore, T., Rose, G., Chubb, J.R., Gaspari, M., Saiardi, A., 2014. Analysis of *Dictyostelium discoideum* inositol pyrophosphate metabolism by gel electrophoresis. *PLoS One* 9, e85533.

Pöggeler, S., 2011. Evolution of Multicopper Oxidase Genes in Coprophilous and Non-Coprophilous Members of the Order Sordariales. *Current Genomics* 12, 95-103.

Pollack, S.J., Atack, J.R., Knowles, M.R., McAllister, G., Ragan, C.I., Baker, R., Fletcher, S.R., Iversen, L.L., Broughton, H.B., 1994. Mechanism of inositol monophosphatase, the putative target of lithium therapy. *Proc Natl Acad Sci U S A* 91, 5766-5770.

Quarmby, L., 2014. Cilia Assembly: A Role for F-Actin in IFT Recruitment. *Current Biology* 24, R796-R798.

Raboy, V., 2001. Seeds for a better future: 'low phytate', grains help to overcome malnutrition and reduce pollution. *Trends in Plant Science* 6, 458-462.

Raboy, V., Young, K.A., Dorsch, J.A., Cook, A., 2001. Genetics and breeding of seed phosphorus and phytic acid. *Journal of Plant Physiology* 158, 489-497.

Rajendran, V., Kalita, P., Shukla, H., Kumar, A., Tripathi, T., 2018. Aminoacyl-tRNA synthetases: Structure, function, and drug discovery. *International Journal of Biological Macromolecules* 111, 400-414.

Raper, C.A., Raper, J.R., 1966. Mutations Modifying Sexual Morphogenesis in Schizophyllum. *Genetics* 54, 1151-1168.

Raper, J.R., Miles, P.G., 1958. The Genetics of *Schizophyllum commune*. *Genetics* 43, 530-546.

Raudaskoski, M., Kothe, E., 2010. Basidiomycete mating type genes and pheromone signaling. *Eukaryot Cell* 9, 847-859.

Raudaskoski, M., Kothe, E., Fowler, T.J., Jung, E.-M., Horton, S.J., 2012. Ras and Rho small G Proteins: Insights from the *Schizophyllum commune* genome sequence and comparisons to other fungi. *Biotechnology and Genetic Engineering Reviews* 28, 61-100.

Reyes, C.L., Rutenber, E., Walter, P., Stroud, R.M., 2007. X-ray structures of the signal recognition particle receptor reveal targeting cycle intermediates. *PLoS One* 2, e607.

Reynolds, T.B., 2009. Strategies for acquiring the phospholipid metabolite inositol in pathogenic bacteria, fungi and protozoa: making it and taking it. *Microbiology (Reading, England)* 155, 1386-1396.

Richter, D.L., Robinson, S.C., Beardslee, M.P., Habarth, M.L., 2008. Differential sensitivity of fungi to lithium chloride in culture media. *Mycol Res* 112, 717-724.

Rigden, D.J., 2008. The histidine phosphatase superfamily: structure and function. *The Biochemical journal* 409, 333-348.

Robert, J.C., Durand, R., 1979. Light and Temperature Requirements during Fruit-Body Development of a Basidiomycete Mushroom, *Coprinus congregates*. *Physiologia Plantarum* 46, 174-178.

Römisch, K., Miller, F.W., Dobberstein, B., High, S., 2006. Human autoantibodies against the 54 kDa protein of the signal recognition particle block function at multiple stages. *Arthritis Research & Therapy* 8, R39-R39.

Roucou, X., Prescott, M., Devenish, R.J., Nagley, P., 2000. A cytochrome c-GFP fusion is not released from mitochondria into the cytoplasm upon expression of Bax in yeast cells. *FEBS letters* 471, 235-239.

Roy, A., Kucukural, A., Zhang, Y., 2010. I-TASSER: a unified platform for automated protein structure and function prediction. *Nature protocols* 5, 725-738.

Sade, Y., Toker, L., Kara, N.Z., Einat, H., Rapoport, S., Moechars, D., Berry, G.T., Bersudsky, Y., Agam, G., 2016. IP3 accumulation and/or inositol depletion: two downstream lithium's effects that may mediate its behavioral and cellular changes. *Translational Psychiatry* 6, e968.

Saiardi, A., 2012a. Cell signalling by inositol pyrophosphates. *Subcell Biochem* 59, 413-443.

Saiardi, A., 2012b. How inositol pyrophosphates control cellular phosphate homeostasis? *Adv Biol Regul* 52, 351-359.

Saiardi, A., 2017. Has Inositol Played Any Role in the Origin of Life? *Life (Basel)* 7.

Saiardi, A., Resnick, A.C., Snowman, A.M., Wendland, B., Snyder, S.H., 2005. Inositol pyrophosphates regulate cell death and telomere length through phosphoinositide 3-kinase-related protein kinases. *Proc Natl Acad Sci U S A* 102, 1911-1914.

Saiardi, A., Sciambi, C., McCaffery, J.M., Wendland, B., Snyder, S.H., 2002. Inositol pyrophosphates regulate endocytic trafficking. *Proc Natl Acad Sci U S A* 99, 14206-14211.

Sampedro, J., Cosgrove, D.J., 2005. The expansin superfamily. *Genome biology* 6, 242.

Sanderfoot, A.A., Raikhel, N.V., 1999. The specificity of vesicle trafficking: coat proteins and SNAREs. *The Plant Cell* 11, 629-642.

Sarkar, S., Floto, R.A., Berger, Z., Imarisio, S., Cordenier, A., Pasco, M., Cook, L.J., Rubinsztein, D.C., 2005. Lithium induces autophagy by inhibiting inositol monophosphatase. *The Journal of Cell Biology* 170, 1101-1111.

Sasakawa, N., Sharif, M., Hanley, M.R., 1995. Metabolism and biological activities of inositol pentakisphosphate and inositol hexakisphosphate. *Biochemical pharmacology* 50, 137-146.

Schubert, D., Raudaskoski, M., Knabe, N., Kothe, E., 2006. Ras GTPase-activating protein gap1 of the homobasidiomycete *Schizophyllum commune* regulates hyphal growth orientation and sexual development. *Eukaryot Cell* 5, 683-695.

Servin, J.A., Campbell, A.J., Borkovich, K.A., 2012. G Protein Signaling Components in Filamentous Fungal Genomes, in: Witzany, G. (Ed.), Biocommunication of Fungi. Springer Netherlands, Dordrecht, pp. 21-38.

Shevchenko, A., Wilm, M., Vorm, O., Mann, M., 1996. Mass spectrometric sequencing of proteins silver-stained polyacrylamide gels. *Anal Chem* 68, 850-858.

Smith, B.E., O'Day, D.H., Proteau, G.A., 1995. Lithium in the mating response and cell cycle of *Saccharomyces cerevisiae*. *Biochemical and biophysical research communications* 206, 401-407.

Sornprasert, R., 1995. Comparison of protein and amino acid in mushroom mycelium and fruiting body *Food* 25(3), 178–184.

Sparvoli, F., Cominelli, E., 2015. Seed Biofortification and Phytic Acid Reduction: A Conflict of Interest for the Plant? *Plants* 4, 728-755.

Stamm, I., Lottspeich, F., Plaga, W., 2005. The pyruvate kinase of *Stigmatella aurantiaca* is an indole binding protein and essential for development. *Mol Microbiol* 56, 1386-1395.

Steggerda, S.M., Black, B.E., Paschal, B.M., 2000. Monoclonal antibodies to NTF2 inhibit nuclear protein import by preventing nuclear translocation of the GTPase Ran. *Mol Biol Cell* 11, 703-719.

Steinberg, G., 2007. Hyphal Growth: a Tale of Motors, Lipids, and the Spitzenkörper. *Eukaryot Cell* 6, 351-360.

Stokes, A.J., Shimoda, L.M., Lee, J.W., Rillero, C., Chang, Y.T., Turner, H., 2006. Fcepsilon RI control of Ras via inositol (1,4,5) trisphosphate 3-kinase and inositol tetrakisphosphate. *Cell Signal* 18, 640-651.

Sun, G.Y., Navidi, M., Yoa, F.G., Lin, T.N., Orth, O.E., Stubbs, E.B., Jr., MacQuarrie, R.A., 1992. Lithium effects on inositol phospholipids and inositol phosphates: evaluation of an in vivo model for assessing polyphosphoinositide turnover in brain. *Journal of neurochemistry* 58, 290-297.

Taylor, S., Wakem, M., Dijkman, G., Alsarraj, M., Nguyen, M., 2010. A practical approach to RT-qPCR-Publishing data that conform to the MIQE guidelines. *Methods* 50, S1-5.

Toledo, A.V., Franco, M.E.E., Yanil Lopez, S.M., Troncozo, M.I., Saparrat, M.C.N., Balatti, P.A., 2017. Melanins in fungi: Types, localization and putative biological roles. *Physiological and Molecular Plant Pathology* 99, 2-6.

Tovar-Herrera, O.E., Batista-García, R.A., Sánchez-Carbente, M.d.R., Iracheta-Cárdenas, M.M., Arévalo-Niño, K., Folch-Mallol, J.L., 2015. A Novel Expansin Protein from the White-Rot Fungus *Schizophyllum commune*. *PLoS ONE* 10, e0122296.

Traxler, L., 2017. Vergleichende Untersuchungen von Inositolmonophosphatase-Überexpressionsmutanten in *Schizophyllum commune*, Friedrich Schiller University Jena.

Tuteja, N., 2009. Signaling through G protein coupled receptors. *Plant Signaling & Behavior* 4, 942-947.

van Impe, K., Hubert, T., De Corte, V., Vanloo, B., Boucherie, C., Vandekerckhove, J., Gettemans, J., 2008. A new role for nuclear transport factor 2 and Ran: nuclear import of CapG. *Traffic* 9, 695-707.

van Peer, A.F., de Bekker, C., Vinck, A., Wösten, H.A.B., Lugones, L.G., 2009. Phleomycin Increases Transformation Efficiency and Promotes Single Integrations in *Schizophyllum commune*. *Applied and Environmental Microbiology* 75, 1243-1247.

Verbsky, J.W., Chang, S.C., Wilson, M.P., Mochizuki, Y., Majerus, P.W., 2005. The pathway for the production of inositol hexakisphosphate in human cells. *J Biol Chem* 280, 1911-1920.

Vidic, I., Berne, S., Drobne, D., Macek, P., Frangez, R., Turk, T., Strus, J., Sepcic, K., 2005. Temporal and spatial expression of ostreolysin during development of the oyster mushroom (*Pleurotus ostreatus*). *Mycol Res* 109, 377-382.

Vivanco, I., Sawyers, C.L., 2002. The phosphatidylinositol 3-Kinase AKT pathway in human cancer. *Nat Rev Cancer* 2, 489-501.

Wessels, J., De Vries, O., Asgeirsdottir, S.A., Schuren, F., 1991. Hydrophobin Genes Involved in Formation of Aerial Hyphae and Fruit Bodies in *Schizophyllum*. *The Plant Cell* 3, 793-799.

Wessels, J.G.H., 1993. Fruiting in the Higher Fungi, in: Rose, A.H. (Ed.), Advances in Microbial Physiology. Academic Press, pp. 147-202.

Wessels, J.G.H., 1994. Developmental Regulation of Fungal Cell Wall Formation. *Annual Review of Phytopathology* 32, 413-437.

Wilson, M.S., Livermore, T.M., Saiardi, A., 2013. Inositol pyrophosphates: between signalling and metabolism. *The Biochemical journal* 452, 369-379.

Wilson, M.S., Saiardi, A., 2017. Importance of Radioactive Labelling to Elucidate Inositol Polyphosphate Signalling. *Top Curr Chem (J)* 375, 14.

Wu, C., 1995. Heat shock transcription factors: structure and regulation. *Annual review of cell and developmental biology* 11, 441-469.

Wyss, M., Brugger, R., Kronenberger, A., Remy, R., Fimbel, R., Oesterhelt, G., Lehmann, M., van Loon, A.P., 1999. Biochemical characterization of fungal phytases (myo-inositol hexakisphosphate phosphohydrolases): catalytic properties. *Appl Environ Microbiol* 65, 367-373.

Xie, N., Ruprich-Robert, G., Chapeland-Leclerc, F., Coppin, E., Lalucque, H., Brun, S., Debuchy, R., Silar, P., 2017. Inositol-phosphate signaling as mediator for growth and sexual reproduction in *Podospora anserina*. *Developmental Biology*.

Xue, C., 2015. Finding the sweet spot: how human fungal pathogens acquire and turn the sugar inositol against their hosts. *mBio* 6, e00109.

Xue, C., Hsueh, Y.-P., Heitman, J., 2008. Magnificent seven: roles of G protein-coupled receptors in extracellular sensing in fungi. *FEMS microbiology reviews* 32, 1010-1032.

Yamagishi, K., Kimura, T., Suzuki, M., Yamaki, K.J., Oita, S., 2005. Identification and overexpression of genes encoding cAMP-dependent protein kinase catalytic subunits in homobasidiomycete *Schizophyllum commune*. *Bioscience, biotechnology, and biochemistry* 69, 2333-2342.

Yang, J., Yan, R., Roy, A., Xu, D., Poisson, J., Zhang, Y., 2015. The I-TASSER Suite: protein structure and function prediction. *Nature methods* 12, 7-8.

Yoon, J.-J., Munir, E., Miyasou, H., Hattori, T., Shimada, M., Terashita, T., 2002. A possible role of the key enzymes of the glyoxylate and gluconeogenesis pathways for fruit-body formation of the wood-rotting basidiomycete *Flammulina velutipes*. *Mycoscience* 43, 327-332.

York, J.D., Odom, A.R., Murphy, R., Ives, E.B., Wente, S.R., 1999. A phospholipase C-dependent inositol polyphosphate kinase pathway required for efficient messenger RNA export. *Science* 285, 96-100.

York, J.D., Ponder, J.W., Majerus, P.W., 1995. Definition of a metal-dependent/$Li^+$-inhibited phosphomonoesterase protein family based upon a conserved three-dimensional core structure. *Proc Natl Acad Sci U S A* 92, 5149-5153.

Young, K.D., 2006. The selective value of bacterial shape. *Microbiology and molecular biology reviews : MMBR* 70, 660-703.

Yuan, J., 2006. Divergence from a dedicated cellular suicide mechanism: exploring the evolution of cell death. *Mol Cell* 23, 1-12.

Zhang J., Yang J., Jang R., Y., Z., 2015. Hybrid structure modeling of G protein-coupled receptors in the human genome. *Submitted.*

Zhang, R.-X., Qin, L.-J., Zhao, D.-G., 2017. Overexpression of the OsIMP Gene Increases the Accumulation of Inositol and Confers Enhanced Cold Tolerance in Tobacco through Modulation of the Antioxidant Enzymes' Activities. *Genes* 8, 179.

Zhang, Y., 2008. I-TASSER server for protein 3D structure prediction. *BMC bioinformatics* 9, 40.

Zhou, M., Li, Y., Hu, Q., Bai, X.-c., Huang, W., Yan, C., Scheres, S.H.W., Shi, Y., 2015a. Atomic structure of the apoptosome: mechanism of cytochrome c- and dATP-mediated activation of Apaf-1. *Genes & development* 29, 2349-2361.

Zhou, X., Liao, W.-J., Liao, J.-M., Liao, P., Lu, H., 2015b. Ribosomal proteins: functions beyond the ribosome. *Journal of Molecular Cell Biology* 7, 92-104.

# 9 Supplemental Material

**Table S1. Proteins involved in inositol phosphate metabolism in human (*Homo sapiens*) and their homologues in *S. commune* H4-8**

| No. | Human Protein | NCBI ID Human | *S. commune* protein homologues | | | | Information |
|---|---|---|---|---|---|---|---|
| | | | schco1 | Protein Identity via BLAST NCBI | schco3 | Protein Identity via BLAST JGI | |
| 1 | Inositol-hexakisphosphate 5-kinase 3 | NP_001136355 | 46826 | Max score 177<br>Total score 177<br>Query cover 94%<br>E-value 3e-51<br>Identity 30% | 2609005 | Score 529<br>E-Value 8.03e-047<br>Identity 40.6%<br>Subject coverage 21.0% | Phosphorylation from Ins 1,2,3,4,5,6-$P_6$ to 5-PP-$IP_5$<br>Phosphorylation from 1-PP-$IP_5$ to $[PP]_2$-$IP_4$ |
| | Inositol-hexakisphosphate 5-kinase 2 | NP_001005909 | 46826 | Max score 172<br>Total score 172<br>Query cover 90%<br>E-value 4e-49<br>Identity 31% | 2609005 | Score 542<br>E-Value 1.27e-048<br>Identity 42.6%<br>Subject coverage 20.4% | |
| | Inositol-hexakisphosphate 5-kinase 1 | NP_001229758 | 46826 | Max score 190<br>Total score 190<br>Query cover 93%<br>E-value 1e-55<br>Identity 31% | 2609005 | Score 559<br>E-Value 9.66e-051<br>Identity 49.1%<br>Subject coverage 18.0% | |
| 2 | Diphosphoinositol pentakisphosphate kinase 2 | NP_001263206 | 63048 | Max score 392<br>Total score 392<br>Query cover 68%<br>E-value 2e-119<br>Identity 34% | 2621615 | Score 1.124<br>E-Value 5.50e-089<br>Identity 50.3%<br>Subject coverage 43.1% | Phosphorylation from Ins 1,2,3,4,5,6-$P_6$ to 1-PP-$InsP_5$<br>Phosphorylation from 5-PP-$InsP_5$ to [PP]2-$InsP_4$ |
| | Diphosphoinositol pentakisphosphate kinase 1 | NP_001124330 | 63048 | Max score 386<br>Total score 386<br>Query cover 59%<br>E-value 2e-116<br>Identity 33% | 2621615 | Score 1.230<br>E-Value 4.17e-093<br>Identity 51.4%<br>Subject coverage 45.3% | |
| 3 | Phospholipase C delta 3 | NP_006216 | 82573 | Max score 301<br>Total score 301<br>Query cover 85%<br>E-value 2e-90<br>Identity 30% | 2628010 | Score 776<br>E-Value 4.74e-065<br>Identity 38.5%<br>Subject coverage 47.3% | PLC catalyses the hydrolysis of membrane PIns-$P_2$, generating the second messengers Ins 1,4,5-$P_3$ and DAG |
| | | - | 79892 | Max score 292<br>Total score 292<br>Query cover 82%<br>E-value 3e-87<br>Identity 3% | 2645684 | Score 758<br>E-Value 4.15e-053<br>Identity 43.1%<br>Subject coverage 36.7% | |
| | | - | 109536 | Max score 145<br>Total score 145<br>Query cover 56%<br>E-value 5e-37<br>Identity 27% | 2669046 | Score 376<br>E-Value 1.13e-031<br>Identity 38.3%<br>Subject coverage 35.5% | |
| | Phospholipase C beta 1 | NP_056007 | - | - | - | - | |
| | Phospholipase C epsilon 1 | NP_057425 | - | - | - | - | |
| | Phospholipase C beta 2 | NP_004564 | - | - | - | - | |
| | Phospholipase C beta 3 | NP_000923 | - | - | - | - | |
| | Phospholipase C beta 4 | NP_877949 | - | - | - | - | |
| | Phospholipase C delta 1 | NP_006216 | 79892 | Max score 301<br>Total score 301<br>Query cover 99%<br>E-value 5e-91<br>Identity 31% | 2645684 | Score 776<br>E-Value 4.74e-065<br>Identity 38.3%<br>Subject coverage 47.3% | |

| | | | | | | | |
|---|---|---|---|---|---|---|---|
| | | | 82573 | Max score 301<br>Total score 301<br>Query cover 97%<br>E-value 4e-91<br>Identity 30% | 2628010 | Score 758<br>E-Value 4.15e-053<br>Identity 43.1%<br>Subject coverage 36.7% | |
| | | | 109536 | Max score 146<br>Total score 146<br>Query cover 60%<br>E-value 3e-37<br>Identity 27% | 2669046 | Score 376<br>E-Value 1.13e-031<br>Identity 38.3%<br>Subject coverage 35.5% | |
| | Phospholipase C gamma 1 | NP_877963 | - | - | - | - | |
| | Phospholipase C gamma 2 | NP_002652 | - | - | - | - | |
| | Phospholipase C delta 4 | NP_116115 | 82573 | Max score 351<br>Total score 351<br>Query cover 97%<br>E-value 1e-109<br>Identity 32% | 2628010 | Score 957<br>E-Value 1.45e-084<br>Identity 40.8%<br>Subject coverage 56.2% | |
| | | | 79892 | Max score 318<br>Total score 318<br>Query cover 97%<br>E-value 2e-97<br>Identity 32% | 2645684 | Score 846<br>E-Value 1.45e-084<br>Identity 40.8%<br>Subject coverage 56.2% | |
| | | | 109536 | Max score 129<br>Total score 129<br>Query cover 60%<br>E-value 9e-32<br>Identity 25% | 2669046 | Score 372<br>E-Value 5.05e-031<br>Identity 39.2%<br>Subject coverage 34.8% | |
| | Phospholipase C zeta 1 | NP_149114 | 82573 | Max score 278<br>Total score 278<br>Query cover 97%<br>E-value 5e-84<br>Identity 31% | 2628010 | Score 755<br>E-Value 9.35e-068<br>Identity 39.2%<br>Subject coverage 34.8% | |
| | | | 79892 | Max score 226<br>Total score 226<br>Query cover 94%<br>E-value 7e-65<br>Identity 27% | 2645684 | Score 613<br>E-Value 2.71e-048<br>Identity 45.2%<br>Subject coverage 27.0% | |
| | | | 109536 | Max score 122<br>Total score 122<br>Query cover 71%<br>E-value 1e-29<br>Identity 25% | 2669046 | Score 428<br>E-Value 5.19e-034<br>Identity 39.3%<br>Subject coverage 42.2% | |
| 4 | Inositol-trisphosphate 3-kinase A | NP_002211 | - | - | - | | Phosphorylation Ins 1,4,5-$P_3$ to Ins 1,3,4,5-$P_4$ |
| | Inositol-trisphosphate 3-kinase B | NP_002212 | - | - | - | | |
| | Inositol-trisphosphate 3-kinase C | NP_079470 | - | - | - | | |
| 5 | Inositol polyphosphate-5-phosphatase J | NP_001271214 | - | - | - | | Dephosphorylation from Ins 1,3,4,5-$P_4$ to Ins 1,3,4-$P_3$<br>Dephosphorylation from Ins 1,4,5-$P_4$ to Ins 1,4-$P_2$ |
| | Inositol polyphosphate-5-phosphatase A | NP_005530 | - | - | - | | |
| | Inositol polyphosphate-5-phosphatase K | NP_057616 | - | - | - | | |
| 6 | Inositol-polyphosphate multikinase | NP_689416 | 70406 | Max score 80.9<br>Total score 80.9<br>Query cover 40%<br>E-value 5e-17<br>Identity 30% | 2639210 | Score 187<br>E-Value 7.26e-009<br>Identity 40%<br>Subject coverage 24.5% | Phosphorylation from Ins 1,3,4,5-$P_4$ to Ins(1,3,4,5,6)$P_5$<br>Phosphorylation from Ins 1,4,5 $P_3$ |

| | | | | | | | |
|---|---|---|---|---|---|---|---|
| | | | | | | | to Ins 1,4,5,6-P$_4$<br>Phosphorylation from Ins 1,4,5,6-P$_4$ to Ins 1,3,4,5,6-P$_5$<br>Phosphorylation from Ins 1,3,4,6-P$_4$ to Ins 1,3,4,5,6-P$_5$ |
| 7 | Inositol-tetrakisphosphate 1-kinase | NP_001136065 | - | - | - | - | Phosphorylation from Ins 1,3,4-P$_3$ to Ins 1,3,4,6-P$_4$ |
| 8 | Inositol polyphosphate-4-phosphatase type I A | NP_001127696 | - | - | - | - | Dephosphorylation from Ins 1,3,4-P$_3$ to Ins 1,3-P$_2$ |
| 9 | Inositol polyphosphate-4-phosphatase type II B | NP_001095139 | - | - | - | - | |
| | Inositol polyphosphate-1-phosphatase | NP_001122400 | - | - | - | - | Dephosphorylation from Ins 1,3,4-P$_3$ to Ins 3,4-P$_2$ |
| 10 | Inositol polyphosphate-5-phosphatase J | NP_001271214 | 72227 | Max score 151<br>Total score 151<br>Query cover 27 %<br>E-value 1e-37<br>Identity 35 % | 2747859 | Score 403<br>E-Value 4.25e-033<br>Identity 41.6%<br>Subject coverage 19.8% | Dephosphorylation from Ins 1,4,5-P$_3$ to Ins 1,4-P$_2$ |
| 11 | | | 79915 | Max score 147<br>Total score 147<br>Query cover 32 %<br>E-value 2e-36<br>Identity 28% | 2645790 | Score 378<br>E-Value 9.07e-030<br>Identity 38.3%<br>Subject coverage 15.3% | |
| 12 | | | 109033 | Max score 108<br>Total score 146<br>Query cover 18 %<br>E-value 3e-24<br>Identity 37% | 2667888 | Score 368<br>E-Value 8.30e-019<br>Identity 46.0%<br>Subject coverage 17.1% | |
| 13 | | | 258354 | Max score 76.6<br>Total score 76.6<br>Query cover 14 %<br>E-value 2e-14<br>Identity 34% | 2638426 | Score 317<br>E-Value 5.18e-012<br>Identity 38.2%<br>Subject coverage 14.2% | |
| 14 | Inositol polyphosphate-5-phosphatase A | NP_005530 | 79915 | Max score 44.7<br>Total score 44.7<br>Query cover 57%<br>E-value 5e-05<br>Identity 23% | - | - | |
| 15 | | | 258354 | Max score 38.1<br>Total score 38.1<br>Query cover 18%<br>E-value 0.005<br>Identity 35% | - | - | |
| 16 | | | 81429 | Max score 37.7<br>Total score 37.7<br>Query cover 40%<br>E-value 0.005<br>Identity 27% | - | - | |
| 17 | Inositol polyphosphate-5-phosphatase K | NP_001271214 | 72227 | Max score 127<br>Total score 127<br>Query cover 72%<br>E-value 2e-31<br>Identity 31% | 2747859 | Score 287<br>E-Value 1.36e-022<br>Identity 38.6%<br>Subject coverage 16.3% | |

| | | | | | | | |
|---|---|---|---|---|---|---|---|
| 18 | | | 79915 | Max score 116<br>Total score 116<br>Query cover 58%<br>E-value 6e-28<br>Identity 29% | 2645790 | Score 348<br>E-Value 1.08e-026<br>Identity 43.3%<br>Subject coverage 12.3% | |
| 19 | | | 81429 | Max score 58.2<br>Total score 58.2<br>Query cover 67%<br>E-value 0.005<br>Identity 25% | - | - | |
| 20 | | | 109033 | Max score 33.9<br>Total score 33.9<br>Query cover 4%<br>E-value 0.12<br>Identity 63% | 2667888 | Score 356<br>E-Value 2.63e-015<br>Identity 41.2%<br>Subject coverage 17.4% | |
| 21 | Myotubularin 1 | NP_000243 | 81454 | Max score 308<br>Total score 308<br>Query cover 89%<br>E-value 2e-94<br>Identity 35% | 2605427 | Score 881<br>E-Value 8.94e-085<br>Identity 53.5%<br>Subject coverage 34.4% | Dephosphorylation from Ins 1,3-$P_2$ to Ins 1-P |
| 22 | Inositol polyphosphate-4-phosphatase type I A | NP_001127696 | - | - | - | - | Dephosphorylation from Ins 3,4 $P_2$ to Ins 3-P |
| 23 | Inositol polyphosphate-1-phosphatase | NP_001122400 | - | - | - | - | Dephosphorylation from Ins 1,4-$P_2$ to Ins 4-P |
| 24 | Inositol monophosphatase 1 | NP_005527 | 47747 | Max score 172<br>Total score 172<br>Query cover 83%<br>E-value 2e-52<br>Identity 39% | 2697390 | Score 450<br>E-Value 2.17e-043<br>Identity 49.4%<br>Subject coverage 52.1% | Dephosphorylation from Ins 1-P to inositol |
| 25 | Inositol monophosphatase 2 | NP_055029 | 47747 | Max score 146<br>Total score 146<br>Query cover 90%<br>E-value 5e-42<br>Identity 34% | 2697390 | Score 353<br>E-Value 1.90e-035<br>Identity 42%<br>Subject coverage 47.6% | Dephosphorylation from Ins 3-P to inositol |
| 26 | Inositol monophosphatase 3 | NP_060283 | 47747 | - | - | - | Dephosphorylation from Ins 4-P to inositol |
| 27 | inositol-pentakisphosphate 2-kinase | NP_073592 | 51281 | Max score 82.4<br>Total score 82.4<br>Query cover 35%<br>E-value 4e-17<br>Identity 31% | 2608225 | Score 230<br>E-Value 3.75e-008<br>Identity 48.9%<br>Subject coverage 17.3% | Phosphorylation from Ins 1,3,4,5,6-$P_5$ to Ins 1,2,3,4,5,6 $P_6$ |

## Table S2. Lithium-induced cytosolic proteins in *S. commune* WT 4-39

| **Spot Number** | **Protein ID schco 3 JGI** | **GO molecular function** | **GO Biological process** | **Interpro Desc** | **KOG GROUP** | **KOG Class** | **KOG Description** | **Fold change** |
|---|---|---|---|---|---|---|---|---|
| 25 | 2023465 | 0005524 ATP binding | | IPR001023 Heat shock protein Hsp70 | Cellular processes and signaling | KOG0100 Posttranslational modification, protein turnover, chaperones | Molecular chaperones HSP105/HSP110/SSE1, HSP70 superfamily | 2.25375 |
| 14 | 2194152 | 0005524 ATP binding | | IPR001023 Heat shock protein Hsp70 | Cellular processes and signaling | KOG0100 Posttranslational modification, protein turnover, chaperones | Molecular chaperones HSP105/HSP110/SSE1, HSP70 superfamily | 3.29309 |

| | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| 3 | 2271574 | 0003824 catalytic activity<br>0008641 small protein activating enzyme activity | 0006464 protein modification<br>0006512 ubiquitin cycle | IPR000594 UBA/THIF-type NAD/FAD binding fold<br>IPR000011 Ubiquitin-activating enzyme, E1 | Cellular processes and signaling | KOG0100 Posttranslational modification, protein turnover, chaperones | Ubiquitin activating enzyme UBA1 | 3.95009 |
| 1 | 65792 | 0008237 metallopeptidase activity<br>0008270 zinc ion binding | 0006508 proteolysis and peptidolysis | IPR014782 Peptidase M1, membrane alanine aminopeptidase, N-terminal<br>IPR006025 Peptidase M, neutral zinc metallopeptidases, zinc-binding site | Metabolism | KOG1184 Amino acid transport and metabolism | Puromycin-sensitive aminopeptidase and related aminopeptidases | 2.34991 |
| 20 | 2552096 | 0003824 catalytic activity<br>0016831 carboxy-lyase activity<br>0000287 magnesium ion binding | | IPR011766 Thiamine pyrophosphate enzyme, C-terminal TPP-binding<br>IPR012110 Pyruvate decarboxylase/ indolepyruvate decarboxylase<br>IPR012000 Thiamine pyrophosphate enzyme, central region | Metabolism | KOG1184 Amino acid transport and metabolism | Thiamine pyrophosphate-requiring enzyme | 2.71125 |
| 74 | 1146700 | 0046872 metal ion binding<br>0004784 superoxide dismutase activity | 0006801 superoxide metabolism | IPR001189 Manganese and iron superoxide dismutase | Metabolism | KOG0876 Inorganic ion transport and metabolism | Manganese superoxide dismutase | 2.71918 |
| 2 | 65792 | 0008237 metallopeptidase activity<br>0008270 zinc ion binding | 0006508 proteolysis and peptidolysis | IPR014782 Peptidase M1, membrane alanine aminopeptidase, N-terminal<br>IPR006025 Peptidase M, neutral zinc metallopeptidases, zinc-binding site | Metabolism | KOG1184 Amino acid transport and metabolism | Puromycin-sensitive aminopeptidase and related aminopeptidases | 3.97762 |
| 29 | 1163199 | | | | Poorly characterized | KOG1995 General function prediction only | Conserved Zn-finger protein | 3.17631 |
| 61 | 1221974 | 0003735 structural constituent of ribosome<br>0008097 5S rRNA binding | 0006412 protein biosynthesis | IPR005484 Ribosomal protein L18/L5<br>IPR005485 Ribosomal protein L5, eukaryotic | Information storage and processing | KOG0875 Translation, ribosomal structure and biogenesis | 60S ribosomal protein L5 | 10.40143 |

**Table S3. Lithium-repressed cytosolic proteins in *S. commune* 4-39**

| Spot Number | Protein ID schco 3 JGI | GO molecular function | GO Biological process | Interpro Desc | KOG GROUP | KOG Class | KOG Description | Fold change |
|---|---|---|---|---|---|---|---|---|
| 34 | 2602263 | 0004497 monooxygenase activity | 0006118 electron transport<br>0006278 RNA dependent DNA replication | IPR002401 Cytochrome P450, E-class, group I<br>IPR001128 Cytochrome P450<br>IPR000477 RNA-directed DNA polymerase (reverse transcriptase)<br>IPR012337 Polynucleotidyl transferase, Ribonuclease H fold | Metabolism | KOG0156 Secondary metabolites biosynthesis, transport and catabolism | Cytochrome P450 CYP2 subfamily | -14.3223 |
| 17 | 2547167 | 0003964 RNA-directed DNA polymerase activity | 0006097 glyoxylate cycle | IPR011076 Malate synthase-like, core<br>IPR001465 Malate synthase<br>IPR006252 Malate synthase A | Metabolism | KOG1261 Energy production and conversion | Malate synthase | -11.3545 |
| 18 | 1129111 | 0003676 nucleic acid binding | 0006097 glyoxylate cycle | IPR011076 Malate synthase-like, core<br>IPR001465 Malate synthase<br>IPR006252 Malate synthase A | Metabolism | KOG1261 Energy production and conversion | Malate synthase | -6.40525 |
| 19 | 1129111 | 0005506 iron ion binding | 0006097 glyoxylate cycle | IPR011076 Malate synthase-like, core<br>IPR001465 Malate synthase<br>IPR006252 Malate synthase A | Metabolism | KOG1261 Energy production and conversion | Malate synthase | -5.71825 |
| 30 | 1088750 | 0020037 heme binding | 0015986 ATP synthesis coupled proton transport | IPR004100 ATPase, F1/V1/A1 complex, alpha/beta subunit, N-terminal<br>IPR000194 ATPase, F1/V1/A1 complex, alpha/beta subunit | Metabolism | KOG1261 Energy production and conversion | F0F1-type ATP synthase, alpha subunit | -4.58648 |

| | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| 15 | 2556130 | 0003723 RNA binding | 0008152 metabolism<br>0006007 glucose catabolism | IPR006124 Metalloenzyme<br>IPR017850 Alkaline-phosphatase-like, core domain<br>IPR011258 BPG-independent PGAM, N-terminal<br>IPR011258 BPG-independent PGAM, N-terminal<br>IPR005995 Phosphoglycerate mutase, 2,3-bisphosphoglycerate-independent | Metabolism | KOG4513 Carbohydrate transport and metabolism | Phosphoglycerate mutase | -3.78238 |
| 24 | 2537897 | 0003824 catalytic activity | 0006561 proline biosynthesis<br>0008152 metabolism | IPR005931 Delta-1-pyrroline-5-carboxylate dehydrogenase 1<br>IPR015590 Aldehyde dehydrogenase<br>IPR016160 Aldehyde dehydrogenase, conserved site<br>IPR016161 Aldehyde/histidinol dehydrogenase | Metabolism | KOG2455 Amino acid transport and metabolism | Delta-1-pyrroline-5-carboxylate dehydrogenase | -3.47657 |
| 68 | 69750 | 0004474 malate synthase activity | 0005975 carbohydrate metabolism | IPR003610 Carbohydrate-binding family V/XII | Metabolism | KOG4513 Carbohydrate transport and metabolism | Candidate chitin-binding protein Carbohydrate-Binding Module Family 12 protein | -3.15293 |
| 67 | 1210187 | 0003824 catalytic activity | 0006096 glycolysis | IPR001576 Phosphoglycerate kinase<br>IPR015911 Phosphoglycerate kinase, conserved site | Metabolism | KOG4513 Carbohydrate transport and metabolism | 3-phosphoglycerate kinase | -2.9708 |
| 41 | 2697237 | 0004474 malate synthase activity | 0006118 electron transport<br>0008152 metabolism | IPR016040 NAD(P)-binding<br>IPR007698 Alanine dehydrogenase/PNT, C-terminal<br>IPR007886 Alanine dehydrogenase/ | Metabolism | KOG2455 Amino acid transport and metabolism | Lysine-ketoglutarate reductase/saccharopine dehydrogenase | -2.96545 |

| | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| | | | | PNT, N-terminal | | | | |
| 9 | 2543807 | 0003824 catalytic activity | 0006118 electron transport<br>0042773 ATP synthesis coupled electron transport | IPR000283 NADH dehydrogenase 75 kDa subunit, conserved site<br>IPR010228 NADH-quinone oxidoreductase, chain G<br>IPR006656 Molybdopterin oxidoreductase<br>IPR001041 Ferredoxin | Metabolism | KOG1261 Energy production and conversion | NADH-ubiquinone oxidoreductase, NDUFS1/75 kDa subunit | -2.6304 |
| 7 | 2520094 | 0004474 malate synthase activity | 0008152 metabolism<br>0006099 tricarboxylic acid cycle | IPR006248 Aconitase, mitochondrial-like<br>IPR001030 Aconitase/3-isopropylmalate dehydratase large subunit, alpha/beta/alpha<br>IPR000573 Aconitase A/isopropylmalate dehydratase small subunit, swivel<br>IPR015928 Aconitase/3-isopropyl malate dehydratase, swivel | Metabolism | KOG0453 Energy production and conversion | Aconitase/homoaconitase (aconitase superfamily) | -2.54895 |
| 54 | 2499212 | 0046961 hydrogen-transporting ATPase activity, rotational mechanism | 0008152 metabolism | IPR005476 Transketolase, C-terminal<br>IPR009014 Transketolase, C-terminal/Pyruvate-ferredoxin oxidoreductase, domain II | Metabolism | KOG0524 Energy production and conversion | Pyruvate dehydrogenase E1, beta subunit | -2.53436 |
| 50 | 62676 | 0015078 hydrogen ion transporter activity | 0008152 metabolism | IPR016040 NAD(P)-binding | Metabolism | KOG0023 Secondary metabolites biosynthesis, transport and catabolism | Alcohol dehydrogenase, class V | -2.43289 |
| 26 | 66738 | 0046933 hydrogen-transporting ATP synthase activity, rotational mechanism | 0006097 glyoxylate cycle<br>0008152 metabolism | IPR000918 Isocitrate lyase and phosphoryl mutase<br>IPR006254 Isocitrate lyase | Metabolism | KOG1260 Energy production and conversion | Isocitrate lyase | -2.24391 |

| | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| 6 | 2721627 | 0016820 hydrolase activity, acting on acid anhydrides, catalyzing transmembrane movement of substances | 0009308 amine metabolism | IPR015798 Copper amine oxidase, C-terminal<br>IPR015800 Copper amine oxidase, N2-terminal<br>IPR015802 Copper amine oxidase, N3-terminal<br>IPR016182 Copper amine oxidase, N-terminal region | Metabolism | KOG1186 Secondary metabolites biosynthesis, transport and catabolism | Copper amine oxidase | -2.10716 |
| 28 | 2703952 | 0003824 catalytic activity | 0008152 metabolism | IPR002618 UTP--glucose-1-phosphate uridylyltransferase | Metabolism | KOG2638 Carbohydrate transport and metabolism | UDP-glucose pyrophosphorylase | -2.09319 |
| 27 | 2704239 | 0030145 manganese ion binding | 0008152 metabolism | IPR005811 ATP-citrate lyase/succinyl-CoA ligase<br>IPR017440 ATP-citrate lyase/succinyl-CoA ligase, active site<br>IPR005810 Succinyl-CoA ligase, alpha subunit<br>IPR016040 NAD(P)-binding | Metabolism | KOG1254 Energy production and conversion | ATP-citrate lyase | -2.0134 |
| 63 | 2536580 | 0004619 phosphoglycerate mutase activity | 0005975 carbohydrate metabolism | IPR005843 Alpha-D-phosphohexomutase, C-terminal<br>IPR005846 Alpha-D-phosphohexomutase, alpha/beta/alpha domain III<br>IPR016055 Alpha-D-phosphohexomutase, alpha/beta/alpha I, II and III | Metabolism | KOG0625 Carbohydrate transport and metabolism | Phosphoglucomutase | -1.97024 |
| 56 | 1205625 | 0046872 metal ion binding | 0008152 metabolism<br>0006007 glucose catabolism | IPR006124 Metalloenzyme<br>IPR017850 Alkaline-phosphatase-like, core domain<br>IPR011258 BPG-independent PGAM, N-terminal | Metabolism | KOG4513 Carbohydrate transport and metabolism | Phospho glycerate mutase | -1.88915 |

| | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| 75 | 1154208 | 0003842 1-pyrroline-5-carboxylate dehydrogenase activity | 0006801 superoxide metabolism | IPR001189 Manganese and iron superoxide dismutase | Metabolism | KOG0876 Inorganic ion transport and metabolism | Manganese superoxide dismutase | -1.79875 |
| 48 | 1129811 | 0016491 oxidoreductase activity | 0006535 cysteine biosynthesis from serine<br>0008152 metabolism | IPR001926 Pyridoxal phosphate-dependent enzyme, beta subunit<br>IPR001216 Cysteine synthase/cystathionine beta-synthase P-phosphate-binding site | Metabolism | KOG1481 Amino acid transport and metabolism | Cysteine synthase | -1.75069 |
| 23 | 2518486 | 0030246 carbohydrate binding | 0008152 metabolism<br>0006007 glucose catabolism | IPR006124 Metalloenzyme<br>IPR017850 Alkaline-phosphatase-like, core domain<br>IPR011258 BPG-independent PGAM, N-terminal<br>IPR005995 Phosphoglycerate mutase, 2,3-bisphosphoglycerate-independent | Metabolism | KOG4513 Carbohydrate transport and metabolism | Phosphoglycerate mutase | -1.67565 |
| 43 | 1115324 | 0004553 hydrolase activity, hydrolyzing O-glycosyl compounds | 0008152 metabolism | IPR016040 NAD(P)-binding<br>IPR002328 Alcohol dehydrogenase, zinc-containing, conserved site | Metabolism | KOG0022 Amino acid transport and metabolism | Lysine-ketoglutarate reductase/saccharopine dehydrogenase | -1.66209 |
| 55 | 2594875 | 0004618 phosphoglycerate kinase activity | 0006096 glycolysis<br>0005975 carbohydrate metabolism<br>0006100 TCA intermediate metabolism<br>0006108 malate metabolism<br>0008152 metabolism | IPR016040 NAD(P)-binding<br>IPR015955 Lactate dehydrogenase/glycoside hydrolase, family 4, C-terminal<br>IPR001252 Malate dehydrogenase, active site<br>IPR001557 L-lactate/ malate dehydrogenase | Metabolism | KOG1494 Energy production and conversion | NAD-dependent malate dehydrogenase | -1.66156 |

| | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| 12 | 2703036 | 0003824 catalytic activity | 0006508 proteolysis and peptidolysis | IPR000994 Peptidase M24, catalytic core<br>IPR000587 Creatinase | Metabolism | KOG2413 Amino acid transport and metabolism | Xaa-Pro aminopeptidase | -1.58595 |
| 5 | 1187071 | 0005488 binding | 0008152 metabolism | IPR005476 Transketolase, C-terminal<br>IPR009014 Transketolase, C-terminal/Pyruvate-ferredoxin oxidoreductase, domain II<br>IPR005478 Bacterial transketolase | Metabolism | KOG4513 Carbohydrate transport and metabolism | Transketolase | -1.58326 |
| 77 | 1306958 | 0016491 oxidoreductase activity | 0006118 electron transport<br>0042773 ATP synthesis coupled electron transport | IPR000283 NADH dehydrogenase 75 kDa subunit, conserved site<br>IPR010228 NADH-quinone oxidoreductase, chain G<br>IPR006656 Molybdopterin oxidoreductase<br>IPR001041 Ferredoxin | Metabolism | KOG1494 Energy production and conversion | NADH-ubiquinone oxidoreductase, NDUFS1/75 kDa subunit | -1.42642 |
| 40 | 2290223 | 0008137 NADH dehydrogenase (ubiquinone) activity | | IPR010109 Citrate synthase, eukaryotic<br>IPR002020 Citrate synthase-like<br>IPR016141 Citrate synthase-like, core | Metabolism | KOG1494 Energy production and conversion | Citrate synthase | -1.41658 |
| 39 | 2512585 | 0016651 oxidoreductase activity, acting on NADH or NADPH | 0008152 metabolism | IPR004790 Isocitrate dehydrogenase NADP-dependent, eukaryotic<br>IPR001804 Isocitrate/isopropylmalate dehydrogenase | Metabolism | KOG1494 Energy production and conversion | NADP-dependent isocitrate dehydrogenase | -1.31632 |
| 8 | 1125316 | 0016491 oxidoreductase activity | 0006118 electron transport<br>0008152 metabolism | IPR016040 NAD(P)-binding<br>IPR001327 Pyridine nucleotide-disulphide oxidoreductase, NAD-binding region | Metabolism | KOG1399 Secondary metabolites biosynthesis, transport and catabolism | Flavin-containing monooxygenase | -1.31109 |

| | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| 37 | 2695306 | 0009055 electron carrier activity | 0006096 glycolysis | IPR001576 Phosphoglycerate kinase<br>IPR015911 Phosphoglycerate kinase, conserved site | Metabolism | KOG1367 Carbohydrate transport and metabolism | 3-phosphoglycerate kinase | -1.31103 |
| 44 | 2697967 | 0003994 aconitate hydratase activity | 0006096 glycolysis | IPR000771 Ketose-bisphosphate aldolase, class-II<br>IPR006411 Fructose-bisphosphate aldolase, class II, yeast/E, coli subtype | Metabolism | KOG4153 Carbohydrate transport and metabolism | Fructose 1,6-bisphosphate aldolase | -1.29034 |
| 69 | 78936 | 0003824 catalytic activity | 0006096 glycolysis<br>0006006 glucose metabolism<br>0008152 metabolism | IPR016040 NAD(P)-binding<br>IPR000173 Glyceraldehyde 3-phosphate dehydrogenase | Metabolism | Carbohydrate transport and metabolism | Glyceraldehyde 3-phosphate dehydrogenase | -1.24638 |
| 21 | 2540966 | 0003824 catalytic activity | | IPR000577 Carbohydrate kinase, FGGY | Metabolism | KOG2517 Carbohydrate transport and metabolism | Ribulose kinase and related carbohydrate kinases | -1.24182 |
| 46 | 1098350 | 0005488 binding | 0008152 metabolism | IPR016040 NAD(P)-binding<br>IPR006140 D-isomer specific 2-hydroxyacid dehydrogenase, NAD-binding<br>IPR006139 D-isomer specific 2-hydroxyacid dehydrogenase, catalytic region | Metabolism | KOG0069 Energy production and conversion | Glyoxylate/hydroxypyruvate reductase (D-isomer-specific 2-hydroxy acid dehydrogenase superfamily) | -1.23176 |
| 35 | 2695306 | 0003824 catalytic activity | 0006096 glycolysis | IPR001576 Phosphoglycerate kinase<br>IPR015911 Phosphoglycerate kinase, conserved site | Metabolism | KOG1367 Carbohydrate transport and metabolism | 3-phosphoglycerate kinase | -1.20469 |
| 22 | 76183 | 0004451 isocitrate lyase activity | 0005975 carbohydrate metabolism | IPR005841 Alpha-D-phosphohexomutase, N-terminal<br>IPR005843 Alpha-D-phosphohexomutase, C-terminal<br>IPR005844 Alpha-D-phosphohexomutase, alpha/beta/alpha domain I | Metabolism | KOG0625 Carbohydrate transport and metabolism | Phosphoglucomutase | -1.15841 |

| | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| 36 | 1135584 | 0005507 copper ion binding | | IPR000560 Histidine acid phosphatase | Metabolism | KOG3720 Lipid transport and metabolism | Lysosomal & prostatic acid phosphatases | -1.15149 |
| 33 | 257792 | 0008131 amine oxidase activity | 0006118 electron transport | IPR010960 Flavocytochrome c<br>IPR003953 Fumarate reductase/succinate dehydrogenase flavoprotein, N-terminal | Metabolism | KOG2404 Energy production and conversion | Fumarate reductase, flavoprotein subunit | -1.14106 |
| 71 | 2695232 | 0048038 quinone binding | 0006241 CTP biosynthesis<br>0006183 GTP biosynthesis<br>0006228 UTP biosynthesis | IPR001564 Nucleoside diphosphate kinase, core | Metabolism | KOG0888 Nucleotide transport and metabolism | Nucleoside diphosphate kinase | -1.13867 |
| 66 | 2254211 | 0016779 nucleotidyl transferase activity | 0006542 glutamine biosynthesis<br>0006807 nitrogen metabolism | IPR008146 Glutamine synthetase, catalytic region<br>IPR008147 Glutamine synthetase, beta-GRasp | Cellular processes and signaling | KOG0683 Amino acid transport and metabolism | Farnesyl cysteine-carboxyl methyltransferase | -3.53493 |
| 72 | 82788 | 0003824 catalytic activity | 0006508 proteolysis and peptidolysis | IPR010259 Proteinase inhibitor I9, subtilisin propeptide<br>IPR000209 Peptidase S8 and S53, subtilisin, kexin, sedolisin | Cellular processes and signaling | KOG1153 Posttranslational modification, protein turnover, chaperones | Subtilisin-related protease/Vacuolar protease B | -2.92896 |
| 73 | 1037839 | 0004775 succinate-CoA ligase (ADP-forming) activity | 0006457 protein folding | IPR001179 Peptidyl-prolyl cis-trans isomerase, FKBP-type | Cellular processes and signaling | KOG0544 Posttranslational modification, protein turnover, chaperones | FKBP-type peptidyl-prolyl cis-trans isomerase | -2.85603 |
| 11 | 1146126 | 0003878 ATP citrate synthase activity | | IPR001023 Heat shock protein Hsp70 | Cellular processes and signaling | KOG0101 Posttranslational modification, protein turnover, chaperones | Molecular chaperones HSP105/HSP110/SSE1, HSP70 superfamily | -2.2163 |
| 42 | 2018351 | 0005488 binding | | IPR001023 Heat shock protein Hsp70 | Cellular processes and signaling | KOG0101 Posttranslational modification, protein turnover, chaperones | Molecular chaperones HSP105/HSP110/SSE1, HSP70 superfamily | -1.90198 |
| 45 | 1038074 | 0046912 transferase activity, transferring acyl groups | 0008152 metabolism | IPR001509 NAD-dependent epimerase/dehydratase | Cellular processes and signaling | KOG1502 Defense mechanisms | Flavonol reductase/cinnamoyl-CoA reductase | -2.1839 |

| | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| 16 | 1146126 | 0016868 intramolecular transferase activity, phosphotransferases | | IPR001023 Heat shock protein Hsp70 | Cellular processes and signaling | KOG0101 Posttranslational modification, protein turnover, chaperones | Molecular chaperones HSP105/HSP110/SSE1, HSP70 superfamily | -1.81774 |
| 13 | 2018348 | 0003824 catalytic activity | | IPR001023 Heat shock protein Hsp70 | Cellular processes and signaling | KOG0101 Posttranslational modification, protein turnover, chaperones | Molecular chaperones HSP105/HSP110/SSE1, HSP70 superfamily | -1.64476 |
| 4 | 1157296 | 0030145 manganese ion binding | | IPR001023 Heat shock protein Hsp70 | Cellular processes and signaling | KOG0103 Posttranslational modification, protein turnover, chaperones | Molecular chaperones HSP105/HSP110/SSE1, HSP70 superfamily | -1.49452 |
| 38 | 1037589 | 0004619 phosphoglycerate mutase activity | | IPR001023 Heat shock protein Hsp71 | Cellular processes and signaling | KOG0101 Posttranslational modification, protein turnover, chaperones | Molecular chaperones HSP105/HSP110/SSE1, HSP70 superfamily | -1.24303 |
| 58 | 1185887 | 0046872 metal ion binding | | IPR000795 Protein synthesis factor, GTP-binding<br>IPR000640 Translation elongation factor EFG/EF2, C-terminal<br>IPR004161 Translation elongation factor EFTu/EF1A, domain 2 | Information storage and processing | KOG0469 Translation, ribosomal structure and biogenesis | Elongation factor 2 | -2.83229 |
| 57 | 2155497 | 0046872 metal ion binding | | IPR000640 Translation elongation factor EFG/EF2, C-terminal<br>IPR004161 Translation elongation factor EFTu/EF1A, domain 2 | Information storage and processing | KOG0469 Translation, ribosomal structure and biogenesis | Elongation factor 2 | -1.31109 |
| 49 | 1208072 | 0004784 superoxide dismutase activity | 0006118 electron transport | IPR004136 2-nitropropane dioxygenase, NPD | | | | -4.29144 |
| 52 | 1194206 | 0003824 catalytic activity | | IPR004000 Actin/actin-like<br>IPR004001 Actin, conserved site | Cellular processes and signaling | KOG0676 Cytoskeleton | Actin and related proteins | -4.11408 |
| 59 | 1107996 | 0030170 pyridoxal phosphate binding | | IPR001395 Aldo/keto reductase | Poorly characterized | KOG1577 General function prediction only | Aldo/keto reductase family proteins | -3.54664 |

| | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| 64 | 1119288 | 0003824 catalytic activity | | IPR001395 Aldo/keto reductase | Poorly characterized | KOG1577 General function prediction only | Aldo/keto reductase family proteins | -2.93462 |
| 53 | 2544070 | 0030145 manganese ion binding | | IPR001395 Aldo/keto reductase | Poorly characterized | KOG1577 General function prediction only | Aldo/keto reductase family proteins | -1.90115 |
| 51 | 2546036 | 0004619 phosphoglycerate mutase activity | | IPR001395 Aldo/keto reductase | Poorly characterized | KOG1577 General function prediction only | Aldo/keto reductase family proteins | -1.89803 |
| 47 | 2703223 | 0046872 metal ion binding | 0008152 metabolism | IPR016040 NAD(P)-binding<br>IPR006140 D-isomer specific 2-hydroxyacid dehydrogenase, NAD-binding<br>IPR006139 D-isomer specific 2-hydroxyacid dehydrogenase, catalytic region | | | | -1.73508 |
| 60 | 1077969 | 0003824 catalytic activity | | IPR002925 Dienelactone hydrolase | | | | -1.63844 |
| 62 | 1213217 | 0005488 binding | 0045454 cell redox homeostasis | IPR005788 Disulphide isomerase<br>IPR006662 Thioredoxin-related<br>IPR011679 Endoplasmic reticulum, protein ERp29, C-terminal | | | | -1.62052 |
| 10 | 258678 | 0016491 oxidoreductase activity | 0006066 alcohol metabolism | IPR000172 Glucose-methanol-choline oxidoreductase, N-terminal<br>IPR007867 Glucose-methanol-choline oxidoreductase, C-terminal<br>IPR012132 Glucose-methanol-choline oxidoreductase | Poorly characterized | KOG1238 General function prediction only | Glucose dehydrogenase/choline dehydrogenase/mandelonitrile lyase (GMC oxidoreductase family) | -1.5383 |
| 65 | 2364242 | 0008270 zinc ion binding | 0006118 electron transport<br>0006979 response to oxidative stress | IPR002016 Haem peroxidase, plant/fungal/bacterial<br>IPR002207 Plant ascorbate peroxidase | | | | -1.40948 |

| | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| 76 | 1039132 | 0003824 catalytic activity | | IPR000504 RNA recognition motif, RNP-1 | | | | -1.31003 |
| 70 | 2145284 | 0016615 malate dehydrogenase activity | | | | | | -1.2808 |
| 31 | 2703952 | 0005488 binding | 0008152 metabolism | IPR002618 UTP--glucose-1-phosphate uridylyltransferase | Poorly characterized | KOG2638 General function prediction only | Conserved Zn-finger protein | -1.26464 |
| 32 | 2366745 | 0016616 oxidoreducta-se activity, acting on the CH-OH group of donors, NAD or NADP as acceptor | 0008152 metabolism | IPR002618 UTP--glucose-1-phosphate uridylyltransferase | Poorly characterized | KOG2638 General function prediction only | Conserved Zn-finger protein | -1.18234 |

## Table S4. Lithium-induced cytosolic proteins in *S. commune* Ras1 active mutant *ras1*$^{G12V}$

| Spot Number | Protein ID schco 3 JGI | GO molecular function | GO Biological process | Interpro Desc | KOG GROUP | KOG Class | KOG Description | Fold change |
|---|---|---|---|---|---|---|---|---|
| 6 | 2535273 | 0003824 catalytic activity<br>0004783 sulfite reductase (NADPH) activity<br>0005506 iron ion binding<br>0009055 electron carrier activity<br>0050661 NADPH binding<br>0020037 heme binding<br>0016491 oxidoreductase activity | 0006118 electron transport<br>0008652 amino acid biosynthesis<br>0008152 metabolism | IPR009014 Transketolase, C-terminal/Pyruvate-ferredoxin oxidoreductase, domain II<br>IPR011786 Sulphite reductase (NADPH) hemoprotein, beta subunit<br>IPR001094 Flavodoxin-like | Metabolism | KOG0560 Inorganic ion transport and metabolism | Sulfite reductase (ferredoxin) | 4.67613 |
| 7 | 2643503 | 0003993 acid phosphatase activity | | IPR000560 Histidine acid phosphatase | Metabolism | KOG3720 Lipid transport and metabolism | Lysosomal & prostatic acid phosphatases | 4.42509 |
| 8 | 2606836 | 0004618 phosphoglycerate kinase activity | 0006096 glycolysis | IPR001576 Phosphoglycerate kinase | Metabolism | KOG1367 Carbohydrate transport and metabolism | 3-phosphoglycerate kinase | 2.0924 |

| | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| 12 | 2632611 | 0003824 catalytic activity<br>0048037 cofactor binding<br>0016616 oxidoreductase activity, acting on the CH-OH group of donors, NAD or NADP as acceptor<br>0005488 binding | 0008152 metabolism | IPR016040 NAD(P)-binding<br>IPR006140 D-isomer specific 2-hydroxy-acid dehydrogenase, NAD-binding<br>IPR006139 D-isomer specific 2-hydroxy-acid dehydrogenase, catalytic region | Metabolism | KOG0069 Energy production and conversion | Glyoxylate/hydroxypyruvate reductase | 2.1364 |
| 14 | 2637783 | 0003824 catalytic activity<br>0016831 carboxy-lyase activity<br>0000287 magnesium ion binding | | IPR011766 Thiamine pyrophosphate enzyme, C-terminal TPP-binding<br>IPR012110 Pyruvate decarboxylase/indolepyruvate decarboxylase | Metabolism | KOG1184 Amino acid transport and metabolism | Thiamine pyrophosphate-requiring enzyme | 3.35103 |
| 15 | 2609880 | 0016616 oxidoreductase activity, acting on the CH-OH group of donors, NAD or NADP as acceptor<br>0004449 isocitrate dehydrogenase (NAD) activity | 0008152 metabolism<br>0006099 tricarboxylic acid cycle | IPR001804 Isocitrate/isopropylmalate dehydrogenase<br>IPR004434 Isocitrate dehydrogenase NAD-dependent, mitochondrial | Metabolism | KOG0785 Amino acid transport and metabolism | Isocitrate dehydrogenase, alpha subunit | 0.473 |
| 16 | 2636349 | 0003824 catalytic activity<br>0005488 binding | 0008152 metabolism | IPR016040 NAD(P)-binding | Metabolism | KOG0023 Secondary metabolites biosynthe-sis, transport and catabolism | Alcohol dehydrogenase, class V | 2.53846 |
| 18 | 2636349 | 0003824 catalytic activity<br>0005488 binding | 0008152 metabolism | IPR016040 NAD(P)-binding | Metabolism | KOG0023 Secondary metabolites biosynthesis, transport and catabolism | Alcohol dehydrogenase, class V | 2.75507 |
| 25 | 2614455 | 0003824 catalytic activity<br>0005488 binding<br>0016491 oxidoreductase activity | 0008152 metabolism | IPR016040 NAD(P)-binding<br>IPR016040 NAD(P)-binding<br>IPR002198 Short-chain dehydrogenase/ | Metabolism | KOG1199 Secondary metabolites biosynthe-sis, transport and catabolism | Short-chain alcohol dehydrogenase/3-hydroxyacyl-CoA dehydrogenase | 2.46948 |

| | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| | | | | reductase SDR<br>IPR002347 Glucose/ribitol dehydrogenase | | | | |
| 29 | 2302125 | 0017111 nucleoside triphosphatase activity<br>0000166 nucleotide binding<br>0042626 ATPase activity, coupled to transmembrane movement of substances<br>0016887 ATPase activity<br>0005524 ATP binding | 0006810 transport | IPR003593 AAA+ ATPase, core<br>IPR011527 ABC transporter, transmembrane region, type 1<br>IPR003439 ABC transporter-like | Metabolism | KOG0054 Secondary metabolites biosynthe-sis, transport and catabolism | Multidrug resistance-associated protein/mitoxantrone resistance protein, ABC superfamily | 2.38802 |
| 33 | 2605124 | 0046872 metal ion binding<br>0004784 superoxide dismutase activity | 0006801 superoxide metabolism | IPR001189 Manganese and iron superoxide dismutase | Metabolism | KOG0876 Inorganic ion transport and metabolism | Manganese superoxide dismutase | 2.9919 |
| 43 | 2532371 | 0004591 oxoglutarate dehydrogenase (lipoamide) activity<br>0016624 oxidoreductase activity, acting on the aldehyde or oxo group of donors, disulfide as acceptor | 0006096 glycolysis<br>0008152 metabolism | IPR011603 2-oxoglutarate dehydrogenase, E1 component<br>IPR001017 Dehydrogenase, E1 component | Metabolism | KOG0450 Carbohydrate transport and metabolism | 2-oxoglutarate dehydrogenase, E1 subunit | 0.47442 |
| 44 | 2643998 | 0003994 aconitate hydratase activity | 0008152 metabolism<br>0006099 tricarboxylic acid cycle | IPR006248 Aconitase, mitochondrial-like<br>IPR001030 Aconitase/3-isopropylmalate dehydrata-se large subunit, alpha/beta/ alpha | Metabolism | KOG0453 Energy production and conversion | Aconitase/homoaconitase (aconitase superfamily) | 2.50894 |

| | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| 55 | 2606654 | 0004550 nucleoside-diphosphate kinase activity<br>0005524 ATP binding | 0006241 CTP biosynthesis<br>0006183 GTP biosynthesis<br>0006228 UTP biosynthesis | IPR000172 Glucose-methanol-choline oxidoreductase, N-terminal<br>IPR012132 Glucose-methanol-choline oxidoreductase | Metabolism | KOG0888 Nucleotide transport and metabolism | Nucleoside diphosphate kinase | 2.4067 |
| 4 | 258678 | 0050660 FAD binding<br>0016614 oxidoreductase activity, acting on CH-OH group of donors | 0006066 alcohol metabolism | IPR000172 Glucose-methanol-choline oxidoreductase, N-terminal<br>IPR012132 Glucose-methanol-choline oxidoreductase<br>IPR007867 Glucose-methanol-choline oxidoreductase, C-terminal | Poorly characterized | KOG1238 General function prediction only | Glucose dehydrogenase/choline dehydrogenase/mandelonitrile lyase (GMC oxidoreductase family) | 2.21066 |
| 26 | 2626705 | | | | | | | 7.73063 |
| 34 | 2751242 | 0030246 carbohydrate binding<br>0004553 hydrolase activity, hydrolyzing O-glycosyl compounds | 0005975 carbohydrate metabolism | IPR003610 Carbohydrate-binding family V/XII | | | | 2.0116 |
| 35 | 2751242 | 0030246 carbohydrate binding<br>0004553 hydrolase activity, hydrolyzing O-glycosyl compounds | 0005975 carbohydrate metabolism | IPR003610 Carbohydrate-binding family V/XII | | | | 9.14523 |
| 22 | 2684323 | 0003924 GTPase activity<br>0005525 GTP binding | | IPR000795 Protein synthesis factor, GTP-binding<br>IPR000640 Translation elongation factor EFG/EF2, C-terminal<br>IPR004161 Translation elongation factor EFTu/EF1A, domain 2 | Information storage and processing | KOG0469 Translation, ribosomal structure and biogenesis | Elongation factor 2 | 2.76409 |

| | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| 54 | 2622068 | 0004540 ribonuclease activity<br>0003723 RNA binding | | IPR001900 Ribonuclease II and R | Information storage and processing | KOG2102 Translation, ribosomal structure and biogenesis | Exosomal 3'-5' exoribonuclease complex, subunit Rrp44/Dis3 | 2.76686 |
| 27 | 2622419 | 0019904 protein domain specific binding | | IPR000308 14-3-3 protein | Cellular processes and signaling | KOG0841 Posttranslational modifica-tion, protein turnover, chaperones | Multifunctional chaperone (14-3-3 family) | 4.23411 |
| 30 | 2147750 | | | | Cellular processes and signaling | KOG0867 Posttranslational modification, protein turnover, chaperones | Glutathione S-transferase | 2.78637 |
| 38 | 1093429 | 0003755 peptidyl-prolyl cis-trans isomerase activity | 0006457 protein folding | IPR002130 Peptidyl-prolyl cis-trans isomerase, cyclophilin-type | Cellular processes and signaling | KOG0865 Posttranslational modification, protein turnover, chaperones | Cyclophilin type peptidyl-prolyl cis-trans isomerase | 2.26861 |
| 39 | 1039708 | 0019787 ubiquitin-like-protein ligase activity | | IPR000608 Ubiquitin-conjugating enzyme, E2 | Cellular processes and signaling | KOG0417 Posttranslational modification, protein turnover, chaperones | Ubiquitin-conjugating enzyme E2 | 2.17841 |
| 40 | 1039708 | 0019787 ubiquitin-like-protein ligase activity | | IPR000608 Ubiquitin-conjugating enzyme, E2 | Cellular processes and signaling | KOG0417 Posttranslational modification, protein turnover, chaperones | Ubiquitin-conjugating enzyme E2 | 3.8466 |
| 46 | 75173 | 0004222 metalloendopeptidase activity | 0006508 proteolysis and peptidolysis | IPR001431 Insulinase-like | Cellular processes and signaling | KOG0960 Posttranslational modification, protein turnover, chaperones | Mitochondrial processing peptidase, beta subunit, and related enzymes (insulinase superfamily) | 2.4897 |

### Table S5. Lithium-repressed cytosolic protein in *S. commune* Ras1 active mutant *ras1*$^{G12V}$

| Spot Number | Protein ID schco 3 JGI | GO molecular function | GO Biological process | Interpro Desc | KOG GROUP | KOG Class | KOG Description | Fold change |
|---|---|---|---|---|---|---|---|---|
| 2 | 257519 | 0003824 catalytic activity<br>0005488 binding | IPR001045 Spermine synthase<br>IPR016040 NAD(P)-binding | IPR001045 Spermine synthase<br>IPR016040 NAD(P)-binding | Metabolism | KOG0172 Amino acid transport and metabolism | Lysine-ketoglutarate reductase/saccharopine dehydrogenase | -2.3412 |
| 5 | 74617 | 0016627 oxidoreductase activity, acting on the CH-CH group of donors<br>0050660 FAD binding<br>0016491 oxidoreductase activity | 0006118 electron transport<br>0006099 tricarboxylic acid cycle | IPR011281 Succinate dehydrogenase, flavoprotein subunit<br>IPR003953 Fumarate reductase/succinate dehydrogenase flavoprotein, N-terminal | Metabolism | KOG2403 Energy production and conversion | Succinate dehydrogenase, flavoprotein subunit | -2.7247 |

| | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| 10 | 68837 | 0003824 catalytic activity<br>0048037 cofactor binding<br>0016616 oxidoreductase activity, acting on the CH-OH group of donors, NAD or NADP as acceptor<br>0005488 binding | 0008152 metabolism | IPR016040 NAD(P)-binding<br>IPR006140 D-isomer specific 2-hydroxyacid dehydrogenase, NAD-binding<br>IPR006139 D-isomer specific 2-hydroxyacid dehydrogenase, catalytic region | Metabolism | KOG0069 Energy production and conversion | Glyoxylate/hydroxypyruvate reductase | -3.5984 |
| 11 | 68837 | 0003824 catalytic activity<br>0048037 cofactor binding<br>0016616 oxidoreductase activity, acting on the CH-OH group of donors, NAD or NADP as acceptor | 0008152 metabolism | IPR016040 NAD(P)-binding<br>IPR006140 D-isomer specific 2-hydroxyacid dehydrogenase, NAD-binding<br>IPR006139 D-isomer specific 2-hydroxyacid dehydrogenase, catalytic region | Metabolism | KOG0069 Energy production and conversion | Glyoxylate/hydroxypyruvate reductase | -2.8174 |
| 13 | 74131 | 0042578 phosphoric ester hydrolase activity | 0005975 carbohydrate metabolism | IPR000146 Inositol phosphatase/fructose-1,6-bisphosphatase | Metabolism | KOG1458 Carbohydrate transport and metabolism | Fructose-1,6-bisphosphatase | -3.5306 |
| 17 | 62676 | 0003824 catalytic activity<br>0005488 binding | 0008152 metabolism | IPR016040 NAD(P)-binding | Metabolism | KOG0023 Secondary metabolites biosynthesis, transport and catabolism | Alcohol dehydrogenase, class V | -2.6038 |
| 19 | 2638699 | 0003824 catalytic activity<br>0004365 glyceraldehyde-3-phosphate dehydrogenase (phosphorylating) activity | 0006096 glycolysis<br>0006006 glucose metabolism<br>0008152 metabolism | IPR016040 NAD(P)-binding<br>IPR000173 Glyceraldehyde 3-phosphate dehydrogenase<br>IPR006424 Glyceraldehyde-3-phosphate dehydrogenase, type I | Metabolism | KOG0657 Carbohydrate transport and metabolism | Glyceraldehyde 3-phosphate dehydrogenase | -3.4612 |
| 21 | 2618819 | 0004639 phosphoribosylaminoimidazole succinocarboxamide synthase activity | 0006164 purine nucleotide biosynthesis | IPR001636 SAICAR synthetase | Metabolism | KOG2835 Nucleotide transport and metabolism | Phosphoribosylamidoimidazole-succinocarboxamide synthase | -4.6317 |

| | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| 45 | 61796 | 0003994 aconitate hydratase activity | 0008152 metabolism<br>0006099 tricarboxylic acid cycle | IPR006248 Aconitase, mitochondrial-like<br>IPR001030 Aconitase/3-isopropylmalate dehydratase large subunit, alpha/beta/alpha<br>IPR000573 Aconitase A/isopropylmalate dehydratase small subunit, swivel<br>IPR015928 Aconitase/3-isopropylmalate dehydratase, swivel | Metabolism | KOG0453 Energy production and conversion | Aconitase/homoaconitase (aconitase superfamily) | -2.7699 |
| 47 | 255314 | 0008237 metallopeptidase activity<br>0005488 binding<br>0008270 zinc ion binding | 0006508 proteolysis and peptidolysis<br>0019370 leukotriene biosynthesis | IPR014782 Peptidase M1, membrane alanine aminopeptidase, N-terminal<br>IPR015211 Peptidase M1, leukotriene A4 hydrolase, aminopeptidase C-terminal<br>IPR006025 Peptidase M, neutral zinc metallopeptidases, zinc-binding site<br>IPR016024 Armadillo-type fold | Metabolism | KOG1047 Amino acid transport and metabolism | Bifunctional leukotriene A4 hydrolase/aminopeptidase LTA4H | -2.7438 |
| 49 | 2600438 | 0016491 oxidoreductase activity | 0008152 metabolism | IPR015590 Aldehyde dehydrogenase<br>IPR016160 Aldehyde dehydrogenase, conserved site<br>IPR016161 Aldehyde/histidinol dehydrogenase | Metabolism | KOG2453 Energy production and conversion | Aldehyde dehydrogenase | -2.3908 |
| 50 | 2637783 | 0003824 catalytic activity<br>0016831 carboxy-lyase activity<br>0000287 magnesium ion binding | | IPR011766 Thiamine pyrophosphate enzyme, C-terminal TPP-binding<br>IPR012110 Pyruvate decarboxylase/indole-pyruvate decarboxylase | Metabolism | KOG1184 Amino acid transport and metabolism | Thiamine pyrophosphate-requiring enzyme | -2.1728 |

| | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| 1 | 2637859 | 0005488 binding | | IPR016024 Armadillo-type fold | Cellular processes and signaling | KOG2005 Posttranslational modification, protein turnover, chaperones | 26S proteasome regulatory complex, subunit RPN1/ PSMD2 | -2.2439 |
| 3 | 2615250 | 0005524 ATP binding | | IPR001023 Heat shock protein Hsp70 | Cellular processes and signaling | KOG0100 Posttranslational modification, protein turnover, chaperones | Molecular chaperones GRP78/BiP/ KAR2, HSP70 superfamily | -2.3707 |
| 9 | 2645649 | 0005515 protein binding<br>0005524 ATP binding | | IPR004000 Actin/actin-like<br>IPR004001 Actin, conserved site | Cellular processes and signaling | KOG0676 Cytoskeleton | Actin and related proteins | -6.2394 |
| 28 | 2624044 | 0046907 intracellular transport | | IPR000156 Ran Binding Protein 1 | Cellular processes and signaling | KOG0864 Intracellular trafficking, secretion, and vesicular transport | Ran-binding protein RANBP1 and related RanBD domain proteins | -2.6433 |
| 31 | 1118835 | 0005524 ATP binding | 0006457 protein folding | IPR003594 ATP-binding region, ATPase-like<br>IPR001404 Heat shock protein Hsp90 | Cellular processes and signaling | KOG0019 Posttranslational modification, protein turnover, chaperones | Molecular chaperone (HSP90 family) | -2.7567 |
| 32 | 2700261 | 0019904 protein domain specific binding | | IPR000308 14-3-3 protein | Cellular processes and signaling | KOG0841 Posttranslational modification, protein turnover, chaperones | Multifunctional chaperone (14-3-3 family) | -6.7100 |
| 48 | 2606458 | 0005524 ATP binding | | IPR001023 Heat shock protein Hsp70 | Cellular processes and signaling | KOG0101 Posttranslational modification, protein turnover, chaperones | Molecular chaperones HSP105/HSP110/SSE1, HSP70 superfamily | -2.3371 |
| 20 | 2632029 | 0003824 catalytic activity | 0008152 metabolism | IPR006992 Amidohydrolase 2 | | | | -2.1192 |
| 24 | 2610865 | 0016491 oxidoreductase activity | | IPR001395 Aldo/keto reductase | Poorly characterized | KOG1577 General function prediction only | Aldo/keto reductase family proteins | -2.2944 |
| 36 | 69750 | 0030246 carbohydrate binding<br>0004553 hydrolase activity, hydrolyzing O-glycosyl compounds | 0005975 carbohydrate metabolism | IPR003610 Carbohydrate-binding family V/XII | | | Candidate chitin-binding protein | -2.6547 |
| 41 | 2699405 | | | | | | | -2.4304 |
| 42 | 2699405 | | | | | | | -2.8509 |
| 51 | 2542709 | | | | | | | -2.3085 |

| | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| 52 | 1177766 | 0003676 nucleic acid binding | | IPR000504 RNA recognition motif, RNP-1 | Poorly characterized | KOG0118 General function prediction only | FOG: RRM domain | -2.1088 |
| 23 | 2622084 | 0003735 structural constituent of ribosome<br>0008097 5S rRNA binding | 0006412 protein biosynthesis | IPR005484 Ribosomal protein L18P/L5E<br>IPR005485 Eukaryotic ribosomal protein L5 | Information storage and processing | KOG0875 Translation, ribosomal structure and biogenesis | 60S ribosomal protein L11 | -2.0711 |
| 37 | 1160443 | 0003743 translation initiation factor activity | 0006413 translational initiation | IPR001884 Eukaryotic initiation factor 5A hypusine (eIF-5A) | Information storage and processing | KOG3271 Translation, ribosomal structure and biogenesis | Translation initiation factor 5A (eIF-5A) | -2.3580 |
| 53 | 2703343 | 0004813 alanine-tRNA ligase activity<br>0016876 ligase activity, forming aminoacyl-tRNA and related compounds<br>0003676 nucleic acid binding<br>0005524 ATP binding | 0006419 alanyl-tRNA aminoacylation<br>0006412 protein biosynthesis | IPR002318 Alanyl-tRNA synthetase, class IIc<br>IPR012947 Threonyl/alanyl tRNA synthetase, SAD<br>IPR003156 Phosphoesterase, DHHA1 | Information storage and processing | Translation, ribosomal structure and biogenesis | Alanyl-tRNA synthetase | -10.996 |

## Table S6. Induced proteins in OEIMP4 in comparison to empty vector control strain (EVC1)

| JGI Schco3 Accession | Folds | Go Desc molecular function | Go Desc Biological Process | Interpro Desc | Cellular component | KOG Group | KOG Class | KOG Desc |
|---|---|---|---|---|---|---|---|---|
| 2598708 | 2.89 | actin binding | | Actin-binding, cofilin/tropomyosin type | intracellular | Cellular processes and signaling | Cytoskeleton | Drebrins and related actin binding proteins |
| 2541047 | 2.69 | microtubule motor activity, ATP binding | microtubule-based movement | Kinesin, motor region | microtubule associated complex | Cellular processes and signaling | Cytoskeleton | Kinesin-like protein |
| 2674409 | 2.33 | | | | | Cellular processes and signaling | Cytoskeleton | Predicted actin-bundling protein |
| 2681330 | 2.31 | catalytic activity, small protein activating enzyme activity | protein modification, ubiquitin cycle, metabolism | UBA/THIF-type NAD/FAD binding fold | | Cellular processes and signaling | Cytoskeleton | Predicted actin-bundling protein |

| | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| 2488292 | 2.97 | | | | | Cellular processes and signaling | Defense mechanisms | Mercaptopyru-vate sulfurtransferase/thiosulfate sulfurtransferase |
| 2687267 | 3.14 | | | | | Cellular processes and signaling | Extracellular structures | SARM protein (with sterile alpha and armadillo motifs) |
| 2704529 | 23.80 | 7S RNA binding | SRP-dependent cotranslational membrane targeting | Signal recognition particle, SRP19 subunit | | Cellular processes and signaling | Intracellular trafficking, secretion, and vesicular transport | Signal recognition particle, subunit Srp19 |
| 2262522 | 23.70 | | protein transport | Snf7 | | Cellular processes and signaling | Intracellular trafficking, secretion, and vesicular transport | Protein involved in glucose derepression and pre-vacuolar endosome protein sorting |
| 2603193 | 21.68 | ATPase activity, coupled to transmembrane movement of substances, ATP binding | transport | ABC transporter, transmembrane region | integral to membrane | Cellular processes and signaling | Intracellular trafficking, secretion, and vesicular transport | Mitochondrial Fe/S cluster exporter, ABC super family |
| 2597192 | 21.66 | DNA binding | DNA methylation | C-5 cytosine-specific DNA methylase | | Cellular processes and signaling | Intracellular trafficking, secretion, and vesicular transport | Golgi transport complex subunit |
| 2678633 | 4.27 | acyltransferase activity, oxidoreductase activity | | Acyltransferase ChoActase/COT/CPT; Aldo/keto reductase | | Cellular processes and signaling | Intracellular trafficking, secretion, and vesicular transport | Vesicle coat complex AP-3, beta subunit |
| 2693248 | 3.49 | 7S RNA binding | SRP-dependent cotranslational membrane targeting | Signal recognition particle, SRP72 subunit, RNA-binding | | Cellular processes and signaling | Intracellular trafficking, secretion, and vesicular transport | Signal recognition particle, subunit Srp72 |
| 2460397 | 3.00 | | protein-mitochondrial targeting, mitochondrial inner membrane protein import | Zinc finger | mitochondrial intermembrane space protein transporter complex | Cellular processes and signaling | Intracellular trafficking, secretion, and vesicular transport | Mitochondrial import inner membrane transloncase |

| | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| 2700443 | 2.96 | binding, ARF guanyl-nucleotide exchange factor activity | | Armadillo-type fold, SEC7-like | Intracellular | Cellular processes and signaling | Intracellular trafficking, secretion, and vesicular transport | Guanine nucleotide exchange factor |
| 1185145 | 2.44 | protein transport | | Snf7 | | Cellular processes and signaling | Intracellular trafficking, secretion, and vesicular transport | Vacuolar assembly/ sorting protein DID2 |
| 2698910 | 2.24 | protein binding, serine-type peptidase activity | cell communication, proteolysis and peptidolysis | Phox-like | | Cellular processes and signaling | Intracellular trafficking, secretion, and vesicular transport | Membrane coat complex Retromer, subunit VPS5/SNX1, Sorting nexins, anrelated PX domain-containing proteins |
| 2678145 | 2.14 | DNA binding, transcription factor activity | regulation of transcription, DNA-dependent | DNA-binding | | Cellular processes and signaling | Intracellular trafficking, secretion, and vesicular transport | Cysteine protease required for autophagy |
| 2007874 | 2.14 | | transport | Nuclear transport factor 2 | Intracellular | Cellular processes and signaling | Intracellular trafficking, secretion, and vesicular transport | Nuclear transport factor 2 |
| 2595664 | 2.12 | protein binding | vesicle-mediated transport | t-SNARE | membrane | Cellular processes and signaling | Intracellular trafficking, secretion, and vesicular transport | SNARE protein Syntaxin 1 and related proteins |
| 2681217 | 2.16 | structural constituent of ribosome, RNA binding | protein biosynthesis | Ribosomal protein S5 | Intracellular, ribosome | Cellular processes and signaling | Nuclear structure | Nucleolar GTPase/ATPase p130 |
| 2288977 | 25.62 | | | | | Cellular processes and signaling | Posttranslational modification, protein turnover, chaperones | Thioredoxin-like protein |
| 2499846 | 23.63 | | | | | Cellular processes and signaling | Posttranslational modification, protein turnover, chaperones | Hsp90 co-chaperone CNS1 (contains TPR repeats) |
| 2696595 | 3.92 | DNA binding | cell redox homeostasis, nucleosome assembly | Histone H4 | nucleosome, nucleus | Cellular processes and signaling | Posttranslational modification, protein turnover, chaperones | Thioredoxin |

| | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| 2164485 | 3.85 | | protein folding | Prefoldin beta-like | prefoldin complex | Cellular processes and signaling | Posttranslational modification, protein turnover, chaperones | Prefoldin subunit 6, KE2 family |
| 2679138 | 3.25 | protein binding, ATP binding | protein folding | Chaperonin Cpn60/TCP-1 | | Cellular processes and signaling | Posttranslational modification, protein turnover, chaperones | Chaperonin complex component, TCP-1 theta subunit (CCT8) |
| 2193905 | 3.21 | antioxidant activity, oxidoreductase activity | | Alkyl hydroperoxide reductase/ Thiol specific antioxidant | | Cellular processes and signaling | Posttranslational modification, protein turnover, chaperones | Alkyl hydroperoxide reductase, thiol specific antioxidant |
| 2675684 | 3.08 | metalloendopeptidase activity, ATP binding, zinc ion binding | protein catabolism, proteolysis and peptidolysis | Peptidase M41; AAA ATPase | membrane | Cellular processes and signaling | Posttranslational modification, protein turnover, chaperones | AAA+-type ATPase containing the peptidase M41 domain |
| 2627563 | 2.91 | | | | | Cellular processes and signaling | Posttranslational modification, protein turnover, chaperones | FKBP-type peptidyl-prolyl cis-trans isomerase |
| 2320137 | 2.56 | peptidyl-prolyl cis-trans isomerase activity | protein folding | Peptidyl-prolyl cis-trans isomerase, cyclophilin-type | | Cellular processes and signaling | Posttranslational modification, protein turnover, chaperones | U-snRNP-associated cyclophilin type peptidyl-prolyl cis-trans isomerase |
| 2695124 | 2.55 | ubiquitin-like-protein ligase activity | | Ubiquitin-conjugating enzyme, E2 | | Cellular processes and signaling | Posttranslational modification, protein turnover, chaperones | Ubiquitin-protein ligase |
| 2600099 | 2.39 | oxidoreductase activity | | Redoxin | | Cellular processes and signaling | Posttranslational modification, protein turnover, chaperones | Alkyl hydroperoxide reductase |
| 2599607 | 2.29 | glutathione peroxidase activity | response to oxidative stress | Glutathione peroxidase | | Cellular processes and signaling | Posttranslational modification, protein turnover, chaperones | Glutathione peroxidase |
| 2747359 | 2.15 | nucleic acid binding, zinc ion binding | | Zinc finger, CCHC-type | | Cellular processes and signaling | Posttranslational modification, protein turnover, chaperones | E3 ubiquitin ligase |

| | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| 2635807 | 2.10 | peptidyl-prolyl cis-trans isomerase activity | protein folding | Peptidyl-prolyl cis-trans isomerase, cyclophilin-type | | Cellular processes and signaling | Posttranslational modification, protein turnover, chaperones | Cyclophilin type peptidyl-prolyl cis-trans isomerase |
| 2213934 | 2.02 | protein folding | | Peptidyl-prolyl cis-trans isomerase, FKBP-type | | Cellular processes and signaling | Posttranslational modification, protein turnover, chaperones | FKBP-type peptidyl-prolyl cis-trans isomerase |
| 2562636 | 2.00 | binding | ubiquitin-dependent protein catabolism | Aspartate decarboxylase-like fold; Ubiquitin fusion degradation protein UFD1 | | Cellular processes and signaling | Posttranslational modification, protein turnover, chaperones | Ubiquitin fusion-degradation protein |
| 2707299 | 22.04 | | | | | Cellular processes and signaling | Signal transduction mechanisms | FOG: FHA domain |
| 2514624 | 3.52 | protein kinase activity, protein-tyrosine kinase activity, protein serine/threonine kinase activity, ATP binding | protein amino acid phosphorylation | Protein kinase, core; Serine/threonine protein kinase-related | | Cellular processes and signaling | Signal transduction mechanisms | p21-activated serine/threonine protein kinase |
| 2022769 | 3.01 | actin binding, binding | cytoskeleton organization and biogenesis | Adenylate cyclase-associated CAP | membrane | Cellular processes and signaling | Signal transduction mechanisms | Adenylate cyclase-associated protein (CAP/Srv2p) |
| 2601695 | 2.85 | | | | | Cellular processes and signaling | Signal transduction mechanisms | Ceramidases |
| 2600833 | 2.79 | mRNA metabolism | mRNA metabolism | Like-Sm ribonucleoprotein, core | ribonucleoprotein complex | Cellular processes and signaling | Signal transduction mechanisms | Sexual differentiation process protein ISP4 |
| 2597350 | 2.59 | catalytic activity | | Histidine triad-like motif | | Cellular processes and signaling | Signal transduction mechanisms | Zinc-binding protein of the histidine triad (HIT) family |
| 2679692 | 24.03 | transcription factor activity, | regulation of transcription | Fungal transcriptional | nucleus | Cellular processes and signaling | Signal transduction mechanisms | ARK protein kinase family |

| | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| | | zinc ion binding | , DNA-dependent | regulatory protein | | | | |
| 2752121 | 23.29 | | | Ubiquitin system component Cue | | Information storage and processing | Chromatin structure and dynamics | SWI-SNF chromatin-remodelling complex protein |
| 2747036 | 2.17 | zinc ion binding | | Zinc finger, MYND-type | | Information storage and processing | Chromatin structure and dynamics | Predicted histone tail methylase containing SET domain |
| 2674751 | 3.45 | DNA binding, endonuclease activity, nuclease activity | DNA repair | AP endonuclease, family 1 | Intracellular | Information storage and processing | Replication, recombination and repair | Apurinic/apyrimidinic endonuclease and related enzymes |
| 2676995 | 2.03 | nucleic acid binding | | Zinc finger, CCCH type | | Information storage and processing | Replication, recombination and repair | Helicases |
| 2665926 | 23.50 | RNA binding | | K Homology, type 1 | | Information storage and processing | RNA processing and modification | Predicted RNA-binding protein, contains KH domains |
| 2603063 | 21.77 | | | | | Information storage and processing | RNA processing and modification | WD40-repeat-containing subunit of the 18S rRNA processing complex |
| 2597573 | 21.12 | hydrolase activity, acting on ester bonds, hydrolase activity | RNA processing, mRNA processing | Lariat debranching enzyme, C-terminal, Metallophosphoesterase, RNA-processing protein, HAT helix | intracellular | Information storage and processing | RNA processing and modification | RNA lariat debranching enzyme |
| 2678057 | 3.12 | | | | | Information storage and processing | RNA processing and modification | Ribosome biogenesis protein - Nop58p/Nop5p |
| 2485735 | 2.99 | | ribosome biogenesis and assembly | Ribosomal protein L7Ae | ribonucleoprotein complex | Information storage and processing | RNA processing and modification | 60S ribosomal protein 15,5kD/SNU13, NHP2/L7 |
| 2563645 | 2.96 | | mRNA processing | Protein of unknown function DUF382 | nucleus | Information storage and processing | RNA processing and modification | Splicing factor 3b, subunit 2 |

| | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| 2595755 | 2.72 | RNA binding | RNA processing, protein modification | SWAP/ Surp | | Information storage and processing | RNA processing and modification | Splicing factor 3a, subunit 1 |
| 2597597 | 2.68 | | | | | Information storage and processing | RNA processing and modification | Polyadenylate-binding protein (RRM super family) |
| 2746442 | 2.46 | RNA binding, nucleic acid binding | mRNA processing | U2 snRNP auxilliary factor, large subunit | nucleus | Information storage and processing | RNA processing and modification | Splicing factor U2AF, large subunit |
| 2266692 | 2.45 | nucleic acid binding, zinc ion binding | | Zinc finger, CCCH-type | | Information storage and processing | RNA processing and modification | Splicing coactivator SRm160/300, subunit SRm160 |
| 2601474 | 2.28 | | protein folding | Peptidyl-prolyl cis-trans isomerase, FKBP-type | | Information storage and processing | RNA processing and modification | RNase P subunit that is not also a subunit of RNase MRP |
| 2488327 | 2.19 | nucleic acid binding, zinc ion binding | | Zinc finger, C2H2-type | nucleus | Information storage and processing | RNA processing and modification | Splicing factor 3a, subunit 2 |
| 2746016 | 2.09 | protein binding | | Leucine-rich repeat | | Information storage and processing | RNA processing and modification | Ran GTPase-activating protein |
| 2601533 | 22.88 | catalytic activity, ATP acid binding, RNA polymerase II transcription factor activity | transcription initiation from Pol II promoter | Transcription Factor IIF, Rap30/Rap74, interaction | transcription factor TFIIF complex | Information storage and processing | Transcription | Transcription initiation factor IIF, small subunit (RAP30) |
| 2694333 | 22.50 | nucleic acid binding | | RNA recognition motif, RNP-1 | | Information storage and processing | Transcription | Nuclear localization sequence binding protein |
| 1157868 | 21.69 | nucleic acid binding, zinc ion binding, RNA polymerase II transcription factor activity | DNA repair, regulation of transcription | Zinc finger, C2H2-type, Ssl1-like | Intracellular, nucleus | Information storage and processing | Transcription | RNA polymerase II transcription initiation/ nucleotide excision repair factor TFIIH, subunit SSL1 |

| | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| 2754015 | 21.17 | positive regulation of transcription | zinc ion binding, positive transcription elongation factor activity | Transcription initiation Spt4 | nucleus | Information storage and processing | Transcription | Transcription elongation factor SPT4 |
| 2679972 | 3.03 | DNA binding, protein binding, transcription factor activity | regulation of transcription, DNA-dependent, regulation of transcription | LAG1, DNA binding | nucleus | Information storage and processing | Transcription | Recombination signal binding protein-J kappa |
| 2029470 | 2.96 | DNA binding | | Lambda repressor-like, DNA-binding | | Information storage and processing | Transcription | Transcription factor MBF1 |
| 2446156 | 2.82 | nucleic acid binding | | RNA recognition motif, RNP-1 | | Information storage and processing | Transcription | Transcription initiation factor TFIID, subunit BDF1 and related bromodomain proteins |
| 2274399 | 2.52 | transcription factor activity, protein dimerization activity | regulation of transcription, DNA-dependent | bZIP transcription factor, bZIP-1 | nucleus | Information storage and processing | Transcription | Transcriptional activator FOSB/c-Fos and related bZIP transcription factors |
| 2539024 | 2.51 | hydrogen ion transporter activity, DNA binding, DNA-directed RNA polymerase activity | transcription | ATPase, F1/V1/A1 complex, alpha/beta subunit, nucleotide-binding; DNA-directed RNA polymerase | proton-transporting two-sector ATPase complex, nucleus | Information storage and processing | Transcription | RNA polymerase I, second largest subunit |
| 2680721 | 2.04 | DNA binding, DNA-directed RNA polymerase activity, ubiquitin-like-protein ligase activity | transcription | DNA-directed RNA polymerase; RNA polymerase Rpb2 | | Information storage and processing | Transcription | RNA polymerase III, second largest subunit |

| | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| 2676225 | 22.02 | structural constituent of ribosome | protein biosynthesis | Ribosomal protein L6 | intracellular, ribosome | Information storage and processing | Translation, ribosomal structure and biogenesis | Mitochondrial/chloroplast ribosomal protein L6 |
| 2527389 | 19.91 | function unknown | | Protein of unknown function UPF0023 | | Information storage and processing | Translation, ribosomal structure and biogenesis | Predicted exosome subunit |
| 2744957 | 5.22 | structural constituent of ribosome, protein biosynthesis | | Ribosomal protein L1 | intracellular, ribosome | Information storage and processing | Translation, ribosomal structure and biogenesis | Mitochondrial/ chloroplast ribosome small subunit |
| 2304861 | 4.18 | | | | | Information storage and processing | Translation, ribosomal structure and biogenesis | Translation initiation factor eIF3, p35 subunit |
| 2641566 | 3.77 | structural constituent of ribosome | protein biosynthesis | Ribosomal protein L7A | intracellular, ribosome | Information storage and processing | Translation, ribosomal structure and biogenesis | 60S ribosomal protein L7A |
| 2613357 | 2.95 | GTPase activity, GTP binding, RNA binding | translational elongation, protein biosynthesis | Protein synthesis factor, GTP-binding; Translation elongation factor EFG/EF2 | | Information storage and processing | Translation, ribosomal structure and biogenesis | Elongation factor-type GTP-binding protein |
| 2595535 | 2.94 | GTPase activity, GTP binding, RNA binding | | Protein synthesis factor, GTP-binding; Translation elongation factor EFG/EF2 | | Information storage and processing | Translation, ribosomal structure and biogenesis | Elongation factor-type GTP-binding protein |
| 2700628 | 2.86 | structural constituent of ribosome | protein biosynthesis | Ribosomal protein L11 | intracellular, ribosome | Information storage and processing | Translation, ribosomal structure and biogenesis | Mitochondrial/ chloroplast ribosomal protein L11 |
| 2247557 | 2.83 | structural constituent of ribosome | protein biosynthesis | Ribosomal protein S21e | intracellular, ribosome | Information storage and processing | Translation, ribosomal structure and biogenesis | 40S ribosomal protein S21 |
| 2663340 | 2.57 | structural constituent of ribosome | protein biosynthesis | Ribosomal protein S16 | intracellular, ribosome | Information storage and processing | Translation, ribosomal structure and biogenesis | Mitochondrial/chloroplast ribosomal protein S16 |

| | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| 2602013 | 2.37 | guanyl nucleotide binding, signal transducer activity | signal transduction, G-protein coupled receptor protein signaling pathway | Guanine nucleotide binding protein (G-protein), alpha subunit | | Information storage and processing | Translation, ribosomal structure and biogenesis | tRNA cytosine-5-methylases and related enzymes of the NOL1/NOP2 |
| 2706403 | 23.63 | translation initiation factor activity, binding | regulation of translational initiation, translational initiation | Translation initiation factor IF2/IF5, eIF4-gamma/eIF5/eIF2-epsilon | | Information storage and processing | Translation, ribosomal structure and biogenesis | Translation initiation factor 5 (eIF-5) |
| 2566607 | 22.95 | RNA binding | | PUA | | Information storage and processing | Translation, ribosomal structure and biogenesis | Ribosome biogenesis protein NIP7 |
| 2707890 | 3.32 | aspartate-tRNA ligase activity, nucleic acid binding, hydrolase activity, acting on acid anhydrides, catalysing transmembrane movement of substances, ATP binding, asparagine-tRNA ligase activity, tRNA ligase activity | proton transport, amino acid activation, aspartyl-tRNA aminoacylation, asparaginyl-tRNA aminoacylation | Aspartyl-tRNA synthetase | proton-transporting two-sector ATPase complex, cytoplasm, membrane | Information storage and processing | Translation, ribosomal structure and biogenesis | Asparaginyl-tRNA synthetase (mitochondrial) |
| 2595295 | 23.23 | ATP binding, nucleotide binding, tRNA ligase activity | amino acid metabolism, amino acid activation, protein biosynthesis | Aminoacyl-tRNA synthetase, class I, conserved site, Asparaginase/glutaminase, Prion protein | cytoplasm, membrane | Metabolism | Amino acid transport and metabolism | Asparaginase |
| 2714397 | 21.33 | carboxy-lyase activity, pyridoxal phosphate binding | carboxylic acid metabolism | Pyridoxal phosphate-dependent decarboxylase | | Metabolism | Amino acid transport and metabolism | Glutamate decarboxylase and related proteins |

| | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| 2599088 | 3.12 | phosphoadenylyl-sulfate reductase (thioredoxin) activity, RNA binding, ribonuclease III activity, transferase activity | RNA processing, sulfate assimilation, phosphoadenylyl sulfate reduction by phosphoadenylyl-sulfate reductase (thioredoxin), cysteine biosynthesis, metabolism | Phosphoadenosine phosphosulphate reductase CysH-type | | Metabolism | Amino acid transport and metabolism | Phosphoadenosine phosphor sulfate reductase |
| 2258133 | 2.97 | | metabolism | Aminotransferase, class V/ Cysteine desulphurase | | Metabolism | Amino acid transport and metabolism | Cysteine desulfurase NFS1 |
| 2535049 | 2.61 | catalytic activity, tryptophan synthase activity, pyridoxal phosphate binding | tryptophan metabolism, metabolism | Pyridoxal phosphate-dependent enzyme, beta subunit; Ribulose-phosphate binding barrel; Tryptophan synthase | | Metabolism | Amino acid transport and metabolism | Tryptophan synthase beta chain |
| 2675838 | 2.04 | asparagine synthase (glutamine-hydrolyzing) activity, nucleic acid binding | asparagine biosynthesis | Asparagine synthase; Prohibitin | membrane | Metabolism | Amino acid transport and metabolism | Asparagine synthase |
| 2619084 | 28.42 | glycerone kinase Activity, ATP binding | glycerol metabolism | Dak kinase, Dak phosphatase, Dihydroxyacetone kinase | | Metabolism | Carbohydrate transport and metabolism | Dihydroxyacetone kinase/ glycerone kinase |
| 2490608 | 26.98 | inositol/ phosphatidylinositol phosphatase activity | | Inositol monophosphatase | | Metabolism | Carbohydrate transport and metabolism | Inositol monophosphatase |
| 2572149 | 22.58 | ribokinase activity | D-ribose metabolism | Ribokinase | | Metabolism | Carbohydrate transport and metabolism | Ribo kinase |
| 2595860 | 2.89 | transporter activity, nucleic acid binding, ubiquitin-like-protein | transport | Major facilitator superfamily | integral to membrane | Metabolism | Carbohydrate transport and metabolism | Permease of the major facilitator superfamily |

| | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| | | ligase activity | | | | | | |
| 2596056 | 2.83 | | | | | Metabolism | Carbohydrate transport and metabolism | Permease of the major facilitator super family |
| 2706632 | 2.62 | carbohydrate binding, chitinase activity | chitin catabolism, carbohydrate metabolism | Carbohydrate-binding family V/XII; Chitinase II | extracellular | Metabolism | Carbohydrate transport and metabolism | Chitinase |
| 2674424 | 2.07 | catalytic activity, binding, oxidoreductase activity | metabolism | NAD(P)-binding | | Metabolism | Carbohydrate transport and metabolism | beta-1,6-N-acetylglucosaminyltransferase, contains WSC domain |
| 2598394 | 2.05 | structural constituent of ribosome, nucleic acid binding, methyl transferase activity | protein biosynthesis | Ribosomal protein L37ae | intracellular, ribosome | Metabolism | Carbohydrate transport and metabolism | Predicted endo-1,3-beta-glucanase |
| 2511993 | 25.04 | | | Rhodanese-like | | Metabolism | Cell cycle control, cell division, chromosome partitioning | M-phase inducer phosphatase |
| 2304724 | 3.71 | guanyl nucleotide binding, signal transducer activity | signal transduction, G-protein coupled receptor protein signaling pathway | Guanine nucleotide binding protein (G-protein), alpha subunit | | Metabolism | Cell cycle control, cell division, chromosome partitioning | G-protein alpha subunit (small G protein super family) |
| 2603681 | 3.28 | nucleic acid binding, zinc ion binding | | Zinc finger, C2H2-type | intracellular | Metabolism | Cell cycle control, cell division, chromosome partitioning | Putative protein methyl transferase involved in meiosis and transcriptional silencing (Dot1) |
| 2491592 | 2.08 | GTP binding | cell cycle | Cell division/ GTP binding protein | | Metabolism | Cell cycle control, cell division, chromosome partitioning | Septin family protein (P-loop GTPase) |
| 2679037 | 2.03 | GTP binding, | cell cycle | Cell division/ | | Metabolism | Cell cycle control, cell | Septin family |

| | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| | | oxidoreductase activity | | GTP binding protein | | | division, chromosome partitioning | protein (P-loop GTPase) |
| 2601430 | 2.03 | catalytic activity, protein kinase activity, protein serine/threonine kinase activity, binding, | protein amino acid phosphorylation, metabolism | NAD(P)-binding; Protein kinase | | Metabolism | Cell cycle control, cell division, chromosome partitioning | Serine/threonine protein kinase Chk2 and related proteins |
| 2170042 | 2.01 | mitochondrial inner membrane protein import | | Zinc finger, Tim10/DDP-type | mitochondrial intermembrane space protein transporter complex | Metabolism | Cell cycle control, cell division, chromosome partitioning | G-protein alpha subunit (small G protein super family) |
| 2571479 | 25.04 | catalytic activity, FAD binding, oxidoreductase | electron transport | FAD-binding, type 2, FAD linked oxidase, N-terminal, Berberine/berberine-like | | Metabolism | Energy production and conversion | Proteins containing the FAD binding domain |
| 2540747 | 23.55 | catalytic activity, FAD binding, oxidoreductase activity | electron transport | FAD-binding, type 2 | | Metabolism | Energy production and conversion | Proteins containing the FAD binding domain |
| 2675630 | 22.87 | oxidoreductase activity, acting on NADH or NADPH | electron transport | mitochondrial inner membrane | ETC complex I subunit | Metabolism | Energy production and conversion | NADH ubiquinone oxidoreductase, NDUFA5/B13 subunit |
| 2575823 | 22.77 | catalytic activity, oxidoreductase activity | electron transport, metabolism | Transketolase, C-terminal/Pyruvate-ferredoxin oxidoreductase, domain II, Flavoprotein pyridine nucleotide cytochrome reductase | | Metabolism | Energy production and conversion | NADP/FAD dependent oxidoreductase |
| 2575012 | 22.03 | electron transporter activity | electron transport | Thioredoxin domain 2 | | Metabolism | Energy production and conversion | ATP binding protein |

| | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| 1178232 | 20.60 | catalytic activity, FAD binding, oxidoreductase activity | electron transport | FAD-binding, type 2, FAD linked oxidase | | Metabolism | Energy production and conversion | Proteins containing the FAD binding domain |
| 2676596 | 3.83 | catalytic activity, carboxyl- and carbamoyl transferase activity, monooxygenase activity, binding, amino acid binding, oxidoreductase activity | electron transport, amino acid metabolism, aromatic compound metabolism, metabolism | NAD(P)-binding; Aspartate/ornithine carbamoyl transferase; Monooxygenase, FAD-binding | | Metabolism | Energy production and conversion | Monooxygenase involved in coenzyme Q (ubiquinone) biosynthesis |
| 2547991 | 3.20 | monooxygenase activity, antioxidant activity, oxidoreductase activity | electron transport, aromatic compound metabolism, metabolism | Monooxygenase, FAD-binding; Alkyl hydroperoxide reductase/Thiol specific antioxidant | | Metabolism | Energy production and conversion | Monooxygenase involved in coenzyme Q (ubiquinone) biosynthesis |
| 2509275 | 3.10 | oxidoreductase activity | | Aldo/keto reductase | | Metabolism | Energy production and conversion | Voltage-gated shaker-like K+ channel |
| 2747681 | 2.96 | oxidoreductase activity | Aldo/keto reductase | | | Metabolism | Energy production and conversion | Voltage-gated shaker-like K+ channel |
| 2293592 | 2.66 | heme binding, iron ion binding, electron carrier activity | electron transport | Cytochrome c, class IA/IB | Mitochondrial electron transport chain | Metabolism | Energy production and conversion | Cytochrome c |
| 2593396 | 2.33 | electron carrier activity | electron transport | Adrenodoxin | | Metabolism | Energy production and conversion | Ferredoxin |
| 2730214 | 2.31 | transition metal ion binding, heme binding | | Cytochrome b5 | integral to membrane | Metabolism | Energy production and conversion | Cytochrome b5 |
| 2597024 | 2.20 | catalytic activity, ATP binding | metabolism | ATP-citrate lyase/succinyl-CoA ligase | | Metabolism | Energy production and conversion | Succinyl-CoA synthetase, beta subunit |

| | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| 2677390 | 26.46 | | | | | Metabolism | Inorganic ion transport and metabolism | TPR-containing nuclear phosphoprotein that regulates K (+) uptake |
| 2598213 | 3.56 | catalytic activity, sulfite reductase (NADPH) activity, iron ion binding, electron carrier activity, NADPH binding, heme binding, oxidoreductase activity | electron transport, amino acid biosynthesis, metabolism | Transketolase; Sulphite reductase (NADPH) hemoprotein; Flavodoxin-like | sulfite reductase complex (NADPH) | Metabolism | Inorganic ion transport and metabolism | Sulfite reductase (ferredoxin) |
| 2509396 | 2.43 | Transferase activity, transferring phosphorrus-containing groups, ATP binding, kinase activity | sulfate assimilation | Adenylyl sulphate kinase | | Metabolism | Inorganic ion transport and metabolism | Adenosine 5'-phosphosulfate kinase |
| 2679539 | 20.55 | hydrolase activity, acting on acid anhydrides, in phosphorus-containing anhydrides, nucleic acid binding, ATP binding, DNA binding, RNA binding | nucleobase, nucleoside, nucleotide and nucleic acid metabolism, nucleotide-excision repair | Xeroderma pigmentosum group D protein, Protein of unknown function DUF1227, Helicase-like, DEXD box c2 type, DNA/RNA helicase | nucleus | Metabolism | Lipid transport and metabolism | Vigilin |
| 2602086 | 3.94 | monooxygenase activity, heme binding, iron ion binding | electron transport | Cytochrome P450 | | Metabolism | Lipid transport and metabolism | Cytochrome P450 CYP4/CYP19/CYP26 subfamilies |

| | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| 2675724 | 3.07 | structural constituent of ribosome, oxidoreductase activity | lipid metabolism, protein biosynthesis | Ribosomal protein L4/L1e; Fatty acid desaturase; Cytochrome b5 | intracellular, ribosome, membrane | Metabolism | Lipid transport and metabolism | Delta 6-fatty acid desaturase / delta-8 sphingolipid desaturase |
| 2116057 | 3.06 | acid phosphatase activity | | Histidine acid phosphatase | | Metabolism | Lipid transport and metabolism | Lysosomal & prostatic acid phosphatases |
| 2681810 | 2.49 | hydrolase activity, oxidoreductase activity | | Metallophosphoesterase | | Metabolism | Lipid transport and metabolism | Acid sphingomyelinase and PHM5 phosphate metabolism protein |
| 2508941 | 2.11 | | lipid metabolism | AB-hydrolase associated lipase region | | Metabolism | Lipid transport and metabolism | Triglyceride lipase-cholesterol esterase |
| 2327633 | 25.61 | | | | | Metabolism | Nucleotide transport and metabolism | Equilibrative nucleoside transportter protein |
| 2700638 | 24.50 | purine-nucleoside phosphorylase activity, transferase activity | nucleobase, nucleoside, nucleotide and nucleic acid metabolism | Inosine guanosine and xanthosine phosphorylase | | Metabolism | Nucleotide transport and metabolism | Purine nucleoside phosphorrylase |
| 2538306 | 24.47 | orotate phosphoribosyltransferase activity, transferase activity | pyrimidine nucleotide biosynthesis, nucleoside metabolism | Orotate phosphoribosyl transferase, Purine/pyrimidine phosphoribosyl transferase | | Metabolism | Nucleotide transport and metabolism | Uridine 5'-monophosphate synthase/orotate phosphoribosyltransferase |
| 1210718 | 22.56 | catalytic activity, hydrolase activity, zinc ion binding | | Cytidine deaminase-like, CMP/dCMP deaminase, zinc-binding | | Metabolism | Nucleotide transport and metabolism | Cytosine deaminase FCY1 and related enzymes |
| 2118550 | 21.25 | catalytic activity | | Glutamine amidotransferase class-I, C-terminal | | Metabolism | Nucleotide transport and metabolism | Predicted glutamine synthetase |
| 2601740 | 3.89 | Nucleobase transportter activity | nucleobase, nucleoside, nucleotide and nucleic acid transport | Permease for cytosine/purines | membrane | Metabolism | Nucleotide transport and metabolism | Uridine permease/thiamine transportter/allantoin transport |

| | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| 2601241 | 23.34 | | electron transport | Cytochrome P450, E-class, group I | | Metabolism | Secondary metabolites biosynthesis, transport and catabolism | Cytochrome P450 CYP2 subfamily |
| 2567671 | 23.27 | nucleic acid binding, ATP binding, zinc ion binding, nucleotide binding, nucleoside triphosphatase activity, ATPase activity | transport | Zinc finger, C2H2-type, ABC transporter-like, CDR ABC transporter, AAA+ ATPase, core | Intracellular, integral to membrane | Metabolism | Secondary metabolites biosynthesis, transport and catabolism | Pleiotropic drug resistance proteins (PDR1-15), ABC superfamily |
| 2595346 | 23.12 | monooxygenase activity, heme binding, iron ion binding | electron transport | Cytochrome P450, E-class, group I | | Metabolism | Secondary metabolites biosynthesis, transport and catabolism | Cytochrome P450 CYP2 subfamily |
| 2270576 | 22.73 | catalytic activity, binding, oxidoreductase activity | metabolism | NAD(P)-binding, Short-chain dehydrogenase/reductase SDR, Glucose/ribitol dehydrogenase | | Metabolism | Secondary metabolites biosynthesis, transport and catabolism | Dehydrogenases with different specificities (related to short-chain alcohol dehydrogenases) |
| 2534031 | 4.98 | O-methyltransferase activity | | O-methyltransferase, family 3 | | Metabolism | Secondary metabolites biosynthesis, transport and catabolism | O-methyltransferase |
| 2582317 | 3.11 | catalytic activity, binding, oxidoreductase activity, zinc ion binding | metabolism | NAD(P)-binding; Alcohol dehydrogenase | | Metabolism | Secondary metabolites biosynthesis, transport and catabolism | Alcohol dehydrogenase, class V |
| 2596825 | 2.48 | isopentenyl-diphosphate delta-isomerase activity, hydrolase activity | isoprenoid biosynthesis | Isopentenyl-diphosphate delta-isomerase | nuclear chromosome | Metabolism | Secondary metabolites biosynthesis, transport and catabolism | Isopentenyl pyrophosphate dimethylallyl pyrophosphate isomerase |
| 2676071 | 2.39 | catalytic activity, ATP binding, glutathione synthase activity | glutathione biosynthesis | PreATP-gRasp-like fold; Glutathione synthase | | Metabolism | Secondary metabolites biosynthesis, transport and catabolism | Glutathione synthetase |

| | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| 2231044 | 25.90 | | | | | Poorly characterized | Function unknown | Uncharacterized conserved protein |
| 2377881 | 23.19 | | | | mitochondrion | Poorly characterized | Function unknown | $Ca^{2+}$-binding transmembrane protein LETM1/MRS7 |
| 2599484 | 23.07 | GTP-binding, nucleic acid binding | | GTP-binding protein, HSR1-related, RNA recognition motif, RNP-1 | intracellular | Poorly characterized | Function unknown | GTPase |
| 2443293 | 2.76 | | | | | Poorly characterized | Function unknown | Uncharacterized conserved protein |
| 2674436 | 2.75 | binding | Armadillo-type fold | | | Poorly characterized | Function unknown | Uncharacterized conserved protein |
| 2678742 | 2.62 | FAD binding, tRNA dihydrouridine synthase activity | tRNA processing | Dihydrouridine synthase, DuS | | Poorly characterized | Function unknown | Uncharacterized conserved protein |
| 2441599 | 2.52 | | | | | Poorly characterized | Function unknown | Uncharacterized conserved protein |
| 2601059 | 2.19 | | | | | Poorly characterized | Function unknown | Uncharacterized conserved protein |
| 2675517 | 24.17 | nucleic acid binding, protein serine/threonine kinase activity, ATP binding, RNA binding | mRNA processing, spliceosome assembly | Tudor, RIO Kinase, Survival motor neuron | nucleus | Poorly characterized | General function prediction only | Serine/threonine protein kinase |
| 2023900 | 24.02 | RNA binding, nucleic acid binding, binding | | RNA recognition motif, RNP-1, Pumilio RNA-binding region | | Poorly characterized | General function prediction only | RNA-binding protein (contains RRM and Pumilio-like repeats) |
| 2055515 | 23.69 | | | | | Poorly characterized | General function prediction only | WD40 repeat-containing protein |

| | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| 2283258 | 23.65 | catalytic activity | | Esterase/lipase/thioesterase | | Poorly characterized | General function prediction only | Predicted alpha/beta hydrolase |
| 2719203 | 23.56 | protein binding, protein kinase activity, protein serine/threonine kinase activity | cell communication, protein amino acid phosphorylation | Phox-like, Protein kinase core, Serine/threonine protein kinase-related, Protein kinas C | | Poorly characterized | General function prediction only | Ribosomal protein S6 kinase and related proteins |
| 2750647 | 23.45 | GTP binding | | GTP1/OBG | | Poorly characterized | General function prediction only | GTP-binding protein CRFG/NOG1 |
| 2698005 | 4.88 | | | | | Poorly characterized | General function prediction only | Predicted Zn-dependent hydrolase (beta-lactamase super family) |
| 2574862 | 4.22 | hydrolase activity | | Beta-lactamase-like | | Poorly characterized | General function prediction only | Predicted metal-dependent hydrolase (beta-lactamase super family) |
| 2677911 | 3.93 | protein kinase activity, protein-tyrosine kinase activity, protein serine/threonine kinase activity | protein amino acid phosphorylation | Protein kinase, core; Serine/threonine protein kinase-related | | Poorly characterized | General function prediction only | Ribosomal protein S6 kinase and related proteins |
| 2682046 | 3.78 | beta-lactamase activity | response to antibiotic, beta-lactam antibiotic catabolism | Beta-lactamase | | Poorly characterized | General function prediction only | FOG: PPR repeat |
| 2033694 | 3.72 | nucleic acid binding, zinc ion binding | | Zinc finger, CCCH-type | | Poorly characterized | General function prediction only | Predicted RNA-binding protein |
| 2680351 | 3.51 | protein binding, antiporter activity, drug transporter activity | multidrug transport | PDZ/DHR/GLGF; Multi antimicrobial extrusion protein MatE | | Poorly characterized | General function prediction only | Uncharacterized membrane protein predicted efflux pump |

| | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| 2699952 | 3.22 | nucleic acid binding, zinc ion binding | | Zinc finger, C2H2-type | intracellular, nucleus | Poorly characterized | General function prediction only | FOG: Zn-finger |
| 2747897 | 3.09 | | | | | Poorly characterized | General function prediction only | FOG: PPR repeat |
| 2600768 | 2.99 | sugar porter activity, trans porter activity | carbohydrate transport, transport | Sugar transporter; General substrate transporter; Major facilitator superfamily | integral to membrane, membrane | Poorly characterized | General function prediction only | Predicted transporter (major facilitator super family) |
| 2542729 | 2.90 | FAD binding, oxidoreductase activity | electron transport | Pyridine nucleotide-disulphide oxidoreductase | | Poorly characterized | General function prediction only | Monodehydroascorbate/ferredoxin reductase |
| 2679069 | 2.79 | GTP binding | protein transport, small GTPase mediated signal transduction | Ras GTPase | | Poorly characterized | General function prediction only | Ras-related small GTPase, Rho type |
| 2677004 | 2.48 | nucleic acid binding, zinc ion binding | | Zinc finger, C2H2-type | intracellular | Poorly characterized | General function prediction only | GTP-binding protein |
| 2680910 | 2.38 | RNA binding, nucleic acid binding | RNA processing | Lupus La protein; RNA recognition motif, RNP-1 | nucleus, ribonucleoprotein complex | Poorly characterized | General function prediction only | FOG: RRM domain |
| 2573839 | 2.22 | GTPase activity, GTP binding | | Dynamin, GTPase region | | Poorly characterized | General function prediction only | Vacuolar sorting protein VPS1, dynamin, and related proteins |
| 2530526 | 2.11 | nucleic acid binding, zinc ion bindig | | Zinc finger, CCCH-type | | Poorly characterized | General function prediction only | CCCH-type Zn-finger protein |
| 2679885 | 2.11 | | | | | Poorly characterized | General function prediction only | Protein involved in autophagocytosis during starvation |
| 2565304 | 2.07 | catalytic activity | | Esterase/lipase/thioesterase | | Poorly characterized | General function prediction only | Esterase D |

| | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| 2257187 | 25.59 | aspartic-type endopeptidase activity, damaged DNA binding | nucleotide-excision repair, protein modification, proteolysis and peptidolysis | Peptidase aspartic, catalytic XPC-binding domain, ubiquitin | | | | |
| 2475239 | 25.43 | ATP binding, nucleotide binding, tRNAse ligase activity, serine-tRNA ligase activity | amino acid activation, protein biosynthesis, seryl-tRNA aminoacylation | Amino acyl-tRNA synthetase, class II (G, H, P and S), Seryl-tRNA synthetase, class IIa, | cytoplasm | | | |
| 2269240 | 25.42 | structural constituent of ribosome | protein biosynthesis | Ribosomal protein S11 | intracellular, ribosome | | | |
| 2596034 | 25.39 | | regulation of signal transduction, response to biotic stimulus | TAP42-like protein | | | | |
| 2681794 | 25.38 | metal ion binding | | Class II aldolase/adducin, N-terminal | | | | |
| 2595836 | 25.23 | catalytic activity, binding, oxidoreductase activity | metabolism | NAD(P)-binding, Short-chain dehydrogenase/reductase SDR, Glucose/ ribitol dehydrogenase | | | | |
| 2094543 | 25.02 | | | | | | | |
| 2501137 | 24.91 | | | | | | | |
| 2615293 | 24.81 | copper ion binding, copper chaperone activity | copper ion transport | Cytochrome C oxidase copper chaperone | mitochondrial intermembrane space | | | |
| 2716936 | 24.74 | monooxygenase activity, peroxidase activity, heme binding, iron ion binding | electron transport, response to oxidative stress | Cytochrome P450, Haem peroxidase | | | | |
| 2598135 | 24.73 | transport ter activity | transport | Major facilitator super family | integral to membrane | | | |

| | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| 2680130 | 24.55 | DNA binding, transcription factor activity, zinc ion binding | transcripttion, regulation of transcription, DNA-dependent | Fungal specific transcription factor | nucleus | | | |
| 2611978 | 24.54 | aminopeptidase activity, prolyl aminopeptidase activity | proteolysis and peptidolysis | Peptidase S33, prolyl aminopeptidase | cytoplasm | | | |
| 2635422 | 24.53 | hydrolase activity, acting on carbon-nitrogen (but not peptide) bonds | | Metal-dependent hydrolase | | | | |
| 2595630 | 24.44 | | | | | | | |
| 2499802 | 24.01 | | | | | | | |
| 2678970 | 23.97 | protein binding, DNA binding, transcription factor regulatory | transcription | Transcription elongation factor, TFIIS/elongin A/ CRSP70, N-terminal | nucleus | | | |
| 2687808 | 23.87 | | | | | | | |
| 2629448 | 23.84 | | | | | | | |
| 2676966 | 23.83 | | | | | | | |
| 2064697 | 23.78 | | | | | | | |
| 2599717 | 23.71 | catalytic activity, binding, oxidoreductase activity | metabolism | NAD(P)-binding | | | | |
| 2681981 | 23.70 | nucleic acid binding | | RNA recognition motif, RNP-1 | | | | |
| 2029060 | 23.65 | hydrolase activity | | NUDIX | | | | |
| 2504962 | 23.60 | | | | | | | |
| 2614441 | 23.59 | catalytic activity | | Esterase/ lipase/thioesterase | | | | |
| 2601028 | 23.55 | catalytic activity, nucleic acid binding, zinc ion binding | metabolism | Amidohydrolase 2, Zinc finger, C2H2-type | Intracelullar | | | |
| 2606239 | 23.46 | | | | | | | |
| 2371381 | 23.45 | | | | | | | |

| | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| 2677729 | 23.39 | | | | | | | |
| 2498088 | 23.38 | zinc ion binding | | Zinc finger, MYND-type | | | | |
| 2596681 | 23.35 | DNA binding | nucleosome assembly | Histone H4 | nucleosome, nucleus | | | |
| 2703628 | 23.35 | monooxygenase activity, heme binding, iron ion binding | | | | | | |
| 2542838 | 23.30 | methyltransferase activity | metabolism | Methyltransferase type 11 | | | | Methyl transferase |
| 2604879 | 23.23 | | | | | | | |
| 2600559 | 23.23 | triacylglycerol lipase activity | lipid metabolism | Lipase, class 3 | | | | |
| 2602824 | 22.98 | catalytic activity, nucleic acid binding, | transport, metabolism | Haloacid dehalogenase-like hydrolase, RNA recognition motif, RNP-1, Nuclear transport factor 2 | intracellular | | | |
| 2596229 | 22.75 | protein binding, binding, ATPase activity, ATP binding, zinc ion binding | | Zinc finger, RING-type, Armadillo-type fold | | | | |
| 2520952 | 22.72 | oxidoreductase activity | | Aldo/ keto reductase | | | | |
| 2680004 | 22.72 | | | | | | | |
| 2678076 | 22.69 | zinc ion binding | | Zinc finger, MYND-type | | | | |
| 2513636 | 22.64 | | | | | | | |
| 2598127 | 22.55 | catalytic activity | | Esterase/lipase/thioesterase | | | | |
| 2680795 | 22.51 | zinc ion binding, binding | | Armadillo-type fold, Zinc finger, MYND-type | | | | |
| 2351474 | 22.49 | protein binding | apoptosis, protein modification | Apoptosis regulator Bcl-2 protein, BAG | | | | |

| | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| 2497117 | 22.45 | | | | | | | |
| 2061457 | 22.45 | | | | | | | |
| 2750188 | 22.44 | DNA binding, transcription factor activity, sigma factor activity | transcription initiation, regulation of transcription (DNA-dependent) | RNA polymerase sigma factor, regions 3 and 4 | | | | |
| 2537421 | 22.39 | protein-nucleus import | | Tetratricopeptide, MLP1/MLP2-like | nuclear pore | | | |
| 2694516 | 22.35 | protein modification | | Ubiquitin | | | | |
| 2233064 | 22.34 | | | | | | | |
| 2505595 | 22.25 | | | | | | | |
| 2027042 | 22.22 | hydrolase activity | | NUDIX hydrolase, core | | | | |
| 2621203 | 21.87 | | | | | | | |
| 2680162 | 21.64 | methyl transferase activity | metabolism | Methyltransferase type 11 | | | | |
| 2596870 | 21.63 | protein binding, transporter activity, zinc ion binding | transport | Zinc finger, PHD-type, Major facilitator superfamily | integral to membrane | | | |
| 2623026 | 21.28 | | | | | | | |
| 2677700 | 21.21 | | | | | | | |
| 2441636 | 20.42 | acid phosphatase activity | Histidine acid phosphatase | | | | | |
| 2675447 | 7.21 | zinc ion binding | | Zinc finger, MYND-type | | | | |
| 2598800 | 5.17 | Metalloendopeptidase activity, metallopeptidase activity, zinc ion binding | proteolysis and peptidolysis | Peptidase M35, deuterolysin | | | | |
| 2685524 | 5.08 | | | | | | | |
| 2677702 | 5.03 | | | | | | | |
| 2639888 | 4.62 | | | | | | | |
| 2699394 | 4.41 | | | | | | | |
| 2629303 | 4.16 | oxidoreductase activity | electron transport | Taurine catabolism dioxygenase | | | | |

| | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| 2598510 | 3.84 | catalytic activity | | Peptidase, trypsin-like serine and cysteine | | | | |
| 2676514 | 3.81 | phosphatase activator activity | | Phosphotyrosyl phosphatase activator | | | | |
| 2680606 | 3.47 | | | | | | | |
| 2521474 | 3.44 | | | | | | | |
| 2508404 | 3.42 | | | | | | | |
| 2703993 | 3.38 | catalytic activity | | Six-hairpin glycosidase-like | | | | |
| 2600993 | 3.29 | | | | | | | |
| 2674711 | 3.07 | hydrolase activity | | Beta-lactamase-like | | | | |
| 2753769 | 2.96 | | | | | | | |
| 2674146 | 2.90 | protein disulfide oxidoreductase activity | cell redox homeostasis, glycerol ether metabolism | Thioredoxin | | | | |
| 2747968 | 2.87 | | | | | | | |
| 2595617 | 2.83 | | | | | | | |
| 2596475 | 2.76 | protein kinase activity, ATP binding, zinc ion binding, transition metal ion binding | electron transport, protein amino acid phosphorylation | Protein kinase, core | | | | |
| 2501340 | 2.72 | protein binding, zinc ion binding | | Zinc finger, RING-type | | | | |
| 2139481 | 2.70 | | | | | | | |
| 2601134 | 2.69 | structural constituent of ribosome, ATP binding, DNA binding, 5-methyltetrahydropteroyltriglutamate-homocysteine S-methyltransferase activity | methionine biosynthesis, protein biosynthesis, DNA topological change, metabolism | Ribosomal protein S6 | Intracellular, ribosome, chromosome | | | |

| | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| 2532725 | 2.66 | oxidoreductase activity, acting on paired donors, with oxidation of a pair of donors resulting in the reduction of molecular oxygen to two molecules of water, oxidoreductase activity | lipid metabolism | Fatty acid desaturase, type 1 | membrane | | | |
| 2470553 | 2.59 | | | | | | | |
| 2214630 | 2.57 | structural constituent of ribosome, RNA binding | protein biosynthesis | Ribosomal protein L21 | Intracellular, ribosome | | | |
| 2585457 | 2.57 | peroxidase activity, heme binding | electron transport, response to oxidative stress | Haem peroxidase, plant/ fungal/ bacterial | | | | |
| 2031430 | 2.56 | | | | | | | |
| 2595602 | 2.54 | catalytic activity, nucleic acid binding, zinc ion binding | | Histidine triad-like motif; Zinc finger, C2H2-type | intracellular | | | |
| 2638568 | 2.53 | | | | | | | |
| 2111247 | 2.52 | copper ion binding | | Cytochrome c oxidase assembly protein CtaG/ Cox11 | | | | |
| 2674593 | 2.51 | guanyl-nucleotide exchange factor activity | small GTPase mediated signal transduction | Guanine-nucleotide dissociation stimulator CDC25 | intracellular | | | |
| 2677737 | 2.48 | phosphorric monoester hydrolase activity, protein tyrosine/ serine/ threonine phosphatase | protein amino acid dephosphorylation, dephosphorylation | Dullard-like phosphatase domain | | | | |

| | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| 2636005 | 2.46 | | | | | | | | |
| 2581083 | 2.46 | RNA binding, nucleic acid binding | mRNA metabolism | Polyadenylate binding protein | | | | | |
| 2743413 | 2.45 | catalytic activity, cyclic-nucleotide phosphodiesterase activity | RNA metabolism | RNA ligase/cyclic nucleotide phosphodiesterase | | | | | |
| 2632297 | 2.42 | RNA binding | | K Homology, type 1 | | | | | |
| 2750677 | 2.36 | | | | | | | | |
| 2751236 | 2.28 | nucleic acid binding | | RNA recognition motif, RNP-1 | | | | | |
| 2675239 | 2.28 | | | | | | | | |
| 2336102 | 2.26 | | | | | | | | |
| 2675127 | 2.26 | ubiquitin-like-protein ligase activity | | Ubiquitin-conjugating enzyme, E2 | | | | | |
| 2696330 | 2.25 | catalytic activity, FAD binding, protein-tyrosine kinase activity, ATP binding | protein amino acid phosphorylation | FAD-linked oxidase | | | | | |
| 2561432 | 2.25 | | | | | | | | |
| 2679184 | 2.20 | zinc ion binding | | Zinc finger, CHY-type | | | | | |
| 2263807 | 2.16 | | | | | | | | |
| 2340383 | 2.14 | | | | | | | | |
| 2596016 | 2.12 | | | GrpE nucleotide exchange factor | | | | | |
| 2597177 | 2.07 | isomerase activity, protein binding, oxidoreductase activity | electron transport | Peptidyl-prolyl cis-trans isomerase | | | | | |
| 2694424 | 2.06 | catalytic activity, binding, oxidoreductase activity | metabolism | NAD(P)-binding | | | | | |

| | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| 2608058 | 2.06 | binding | | Armadillo-type fold | | | | |
| 2702672 | 2.05 | protein binding, hydroxymethylglutaryl-CoA reductase (NADPH) activity, coenzyme binding, coenzyme A metabolism | | BTB/POZ | | | | |
| 2570260 | 2.04 | catalytic activity, DNA binding, ATP binding, nuclease activity | DNA repair, metabolism | Helix-hairpin-helix motif; Carbamoyl phosphate synthetase | nucleus | | | |
| 2675514 | 2.02 | | | | | | | |
| 2435065 | 2.01 | | | | | | | |

## Table S7. Repressed proteins in OEIMP4 compared to the empty vector control strain (EVC1)

| **JGI Schco3 Accession** | **Folds** | **Go Desc molecular function** | **Go Desc Biological Process** | **Interpro Desc** | **Cellular component** | **KOG Group** | **KOG Class** | **KOG Desc** |
|---|---|---|---|---|---|---|---|---|
| 2563832 | -25.32 | Metallopeptidase activity, protein dimerization activity, hydrolase activity | proteolysis and peptidelysis | Peptidase M20 | | Metabolism | Amino acid transport and metabolism | Aminoacylase ACY1 and related metalloexopeptidase |
| 2468941 | -3.43 | oxidoreductase activity, acting on the aldehyde or oxo group of donors, disulfide as acceptor; oxoglutarate dehydrogenase (lipoamide) activity | glycolysis, proteolysis and peptidolysis, metabolism | Dehydrogenase, E1 | membrane | Metabolism | Carbohydrate transport and metabolism | Predicted 2-oxoglutarate dehydrogenase, E1 subunit |
| 2030611 | -21.13 | hydrolase activity, hydrolyzing O-glycosyl compounds | carbohydrate metabolism | Glycoside hydrolase, family 1 | | Metabolism | Carbohydrate transport and metabolism | Beta-glucosidase |

| | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| 2481654 | -21.48 | hydrolase activity, hydrolyzing O-glycosyl compounds | carbohydrate metabolism | Glycoside hydrolase, family 1 | | Metabolism | Carbohydrate transport and metabolism | Beta-glucosidase |
| 2505453 | -23.52 | glycerone kinase activity | glycerol metabolism | Dak Kinase | | Metabolism | Carbohydrate transport and metabolism | Dihydroxyacetone kinase/ glycerone kinase |
| 2729808 | -27.85 | carbohydrate binding, catalytic activity, isomerase activity | carbohydrate metabolism | Glycoside hydrolase-type carbohydrate-binding | | Metabolism | Carbohydrate transport and metabolism | Predicted mutarotase |
| 2677621 | -23.89 | DNA binding, RNA polymerase II transcription factor activity | transcription initiation | Histone-fold | transcription factor complex | Metabolism | Cell cycle control, cell division, chromosome partitioning | Transcription-associated recombination protein - Thp1p |
| 2493976 | -22.09 | | | Gpi16 subunit, GPI transamidase component | integral to membrane, GPI-anchor transamidase complex | Cellular processes and signaling | Cell wall/ membrane/ envelope biogenesis | GPI transamidase complex, GPI16/ PIG-T component, involved in glycosyl phosphatidylinositol anchor biosynthesis |
| 2677610 | -3.96 | | | | | Information storage and processing | Chromatin structure and dynamics | SWI-SNF chromatin-remodeling complex protein |
| 2674879 | -23.37 | | | BRCT | intracellular | Information storage and processing | Chromatin structure and dynamics | Acinus |
| 2680166 | -3.30 | phosphomethylpyrimidine kinase activity | thiamin biosynthesis | Phosphomethyl-pyrimidine kinase type-2 | | Metabolism | Coenzyme transport and metabolism | Phosphomethylpyrimidine kinase |
| 2677987 | -22.19 | phosphoric monoester hydrolase activity, protein tyrosine/ serine/ threonine phosphatase activity, protein tyrosine phosphatase activity | protein amino acid dephosphorylation, dephosphorylation | Protein-tyrosine phosphatase | | Cellular processes and signaling | Defense mechanisms | Protein tyrosine phosphatase CDC14 |

| | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| 2034011 | -2.23 | disulfide oxidoreductase activity | electron transport | FAD-dependent pyridine nucleotide | | Metabolism | Energy production and conversion | Sulfide quinone oxidoreductase/ flavo binding protein |
| 2257982 | -3.29 | transition metal ion binding, heme binding | | Cytochrome b5 | | Metabolism | Energy production and conversion | Cytochrome b5 |
| 2595828 | -24.05 | monooxygenase activity, oxidoreductase activity | electron transport, aromatic compound metabolism, metabolism | Monooxygenase, FAD-binding; Aromatic-ring hydroxylase | | Metabolism | Energy production and conversion | Kynurenine 3-monooxygenase and related flavoprotein monooxygenases |
| 2579775 | -24.56 | binding | transport | Mitochondrial carrier protein | Mitochondrial inner membrane, membrane | Metabolism | Energy production and conversion | Predicted mitochondrial carrier protein |
| 2573262 | -2.09 | | | | | | function unknown | Protein of unknown function DUF323 |
| 2540553 | -2.11 | | | | | | function unknown | Predicted membrane protein |
| 2679299 | -2.53 | protein-glutamine gamma-glutamyl-transferase activity | peptide cross-linking | Transglutaminase, C-terminal | | Poorly characterized | Function unknown | Uncharacterized conserved protein |
| 2676154 | -2.97 | | | | | Poorly characterized | Function unknown | Conserved WD40 repeat-containing protein |
| 2565102 | -22.38 | | | Transmembrane receptor, eukaryote | integral to membrane | Poorly characterized | Function unknown | Predicted membrane protein |
| 2498149 | -2.21 | oxidoreductase activity, acting on the CH-CH group of donors, acyl-CoA dehydrogenase activity | electron transport, metabolism | Acyl-CoA oxidase/dehydrogenase, type 1 | | Poorly characterized | General function prediction only | Predicted acyl-CoA dehydrogenase |
| 1126608 | -2.22 | sugar porter activity, transporter activity | Carbohydrate transport, transport | Sugar transporter, General substrate transporter | integral to membrane, membrane | Poorly characterized | General function prediction only | Predicted transporter (major facilitator superfamily) |

| | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| 2680465 | -2.46 | catalytic activity | | Esterase/ lipase/ thioesterase | | Poorly characterized | General function prediction only | Carboxylesterase and related proteins |
| 2602357 | -2.75 | protein binding | amino acid transport, transport | BTB/POZ fold, Amino acid permease, conserved site | integral to membrane, membrane | Poorly characterized | General function prediction only | Uncharacterized conserved protein |
| 2695406 | -3.00 | | | | | Poorly characterized | General function prediction only | Predicted transporter (major facilitator superfamily) |
| 2319811 | -3.41 | sugar porter activity, transporter activity | carbohydrate transport, transport | Sugar transporter | integral to membrane, membrane | Poorly characterized | General function prediction only | Predicted transporter (major facilitator superfamily) |
| 2749685 | -3.89 | sugar porter activity, transporter activity | carbohydrate transport, transport | Sugar transporter | integral to membrane, membrane | Poorly characterized | General function prediction only | Predicted transporter (major facilitator superfamily) |
| 2362924 | -4.37 | transporter activity | transport | Major facilitator superfamily | integral to membrane | Poorly characterized | General function prediction only | Synaptic vesicle transporter SVOP and related transporters (major facilitator superfamily) |
| 2634498 | -22.73 | | | | | Poorly characterized | General function prediction only | Predicted thioesterase |
| 2688026 | -22.85 | sugar porter activity, transporter activity | carbohydrate transport, transport | Sugar transporter; General substrate transporter; Major facilitator superfamily | integral to membrane, membrane | Poorly characterized | General function prediction only | Predicted transporter (major facilitator super family) |
| 2174709 | -23.04 | sugar porter activity, transporter activity | carbohydrate transport, transport | Sugar transporter | integral to membrane, membrane | Poorly characterized | General function prediction only | Predicted transporter (major facilitator super family) |
| 2696574 | -23.14 | GTP binding, sugar porter activity, transporter activity | carbohydrate transport, transport | GTP1/OBG; GTP-binding protein; Sugar transporter | Intracellular, membrane, integral to membrane | Poorly characterized | General function prediction only | Predicted transporter (major facilitator super family) |

| | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| 2034237 | -23.71 | sugar porter activity, transporter activity | carbohydrate transport, transport | Sugar transporter | integral to membrane, membrane | Poorly characterized | General function prediction only | Predicted transporter (major facilitator super family) |
| 2464691 | -23.97 | sugar porter activity, transporter activity | carbohydrate transport, transport | Sugar transporter | integral to membrane, membrane | Poorly characterized | General function prediction only | Predicted transporter (major facilitator super family) |
| 2675908 | -24.22 | sugar porter activity, transporter activity | carbohydrate transport, transport | Sugar transporter | integral to membrane, membrane | Poorly characterized | General function prediction only | Predicted transporter (major facilitator super family) |
| 2043563 | -24.76 | sugar porter activity, transporter activity | carbohydrate transport, transport | Sugar transporter | integral to membrane, membrane | Poorly characterized | General function prediction only | Predicted transporter (major facilitator super family) |
| 2622593 | -25.87 | sugar porter activity, transporter activity | carbohydrate transport, transport | Sugar transporter | integral to membrane, membrane | Poorly characterized | General function prediction only | Predicted transporter (major facilitator super family) |
| 2682492 | -23.20 | transporter activity | transport | Major facilitator super family | integral to membrane, membrane | Poorly characterized | General function prediction only | Synaptic vesicle transporter SVOP and related transporters |
| 2486786 | -2.16 | | | | | Metabolism | Inorganic ion transport and metabolism | Acetyl-CoA transporter |
| 2368187 | -2.47 | cation transporter activity | cation transport | Cation efflux protein | membrane | Metabolism | Inorganic ion transport and metabolism | Mitochondrial Fe2+ transporter MMT1 and related transporters (cation diffusion facilitator super family) |
| 2490039 | -21.77 | cation transporter activity | cation transport | Cation efflux protein | membrane | Metabolism | Inorganic ion transport and metabolism | $Zn^{2+}$ transporter ZNT1 and related $Cd^{2+}/Zn^{2+}$ transporters (cation diffusion facilitator superfamily |

| | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| 2696284 | -2.25 | peroxisome division | Peroxisomal membrane | Peroxisomal biogenesis factor 11 | | Cellular processes and signaling | Intracellular trafficking, secretion, and vesicular transport | Peroxisomal biogenesis protein (peroxin) |
| 2632335 | -2.07 | catalytic activity, monooxygenase activity, heme binding, binding, iron ion binding | electron transport, metabolism | NAD(P)-binding, Cytochrome P450 | | Metabolism | Lipid transport and metabolism | Cytochrome P450 CYP4/ CYP19/ CYP26 subfamilies |
| 2473616 | -2.16 | oxidoreductase activity, acting on the CH-CH group of donors, acyl-CoA dehydrogenase activity | electron transport, metabolism | Acyl-CoA oxidase/ dehydrogenase, type 1 | | Metabolism | Lipid transport and metabolism | Very-long-chain acyl-CoA dehydrogenase |
| 2316010 | -20.79 | catalytic activity, 3',5'-cyclic-nucleotide phosphodiesterase activity | signal transduction, metabolism | Sterol desaturase | | Metabolism | Lipid transport and metabolism | C-4 sterol methyl oxidase |
| 2746430 | -21.80 | catalytic activity, lyase activity | metabolism | Prenyltransferase/ squalene oxidase, Terpene synthase | | Metabolism | Lipid transport and metabolism | Oxidosqualene-lanosterol cyclase and related proteins |
| 2511921 | -22.01 | iron ion binding; oxidoreductase activity, acting on paired donors | lipid metabolism, fatty acid biosynthesis | Fatty acid desaturase, type 1 | endoplasmic reticulum, membrane | Metabolism | Lipid transport and metabolism | Fatty acid desaturase |
| 2550250 | -22.13 | iron ion binding; oxidoreductase activity | lipid metabolism, fatty acid biosynthesis | Fatty acid desaturase, type 1 | endoplasmic reticulum, membrane | Metabolism | Lipid transport and metabolism | Fatty acid desatuase |
| 2035580 | -22.51 | catalytic activity | metabolism | Crotonase, core | | Metabolism | Lipid transport and metabolism | Enoyl-CoA hydratase/ isomerase |
| 1130394 | -2.47 | hydrolase activity, acting on carbon-nitrogen | urea metabolism, nitrogen metabolism | Metal-dependent hydrolase, Amidohydrolase 1, Urease | | Metabolism | Nucleotide transport and metabolism | Dihydroorotase and related enzymes |

| | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| 2314901 | -24.25 | ribonucleoside-diphosphate reductase activity, transition metal ion binding, oxidoreductase activity | deoxyribonucleoside diphosphate metabolism | Ribonucleotide reductase | | Metabolism | Nucleotide transport and metabolism | Ribonucleotide reductase, beta subunit |
| 2597700 | -2.29 | nucleoside triphosphatase activity, nucleotide binding | protein catabolism | AAA+ ATPase, core, 26S proteasome subunit P45 | cytoplasm, nucleus | Cellular processes and signaling | Posttranslational modification, protein turnover, chaperones | 26S proteasome regulatory complex, ATPase RPT5 |
| 2603157 | -2.45 | aspartic-type endopeptidase activity | proteolysis and peptidolysis | Peptidase aspartic, active site | membrane | Cellular processes and signaling | Posttranslational modification, protein turnover, chaperones | Aspartyl protease |
| 2623405 | -2.72 | peptidase activity | proteolysis and peptidolysis | Peptidase M28 | | Cellular processes and signaling | Posttranslational modification, protein turnover, chaperones | Transferrin receptor and related proteins containing the protease-associated domain |
| 1148905 | -20.50 | protein binding, zinc ion binding | | Zinc finger, RING-type | | Cellular processes and signaling | Posttranslational modification, protein turnover, chaperones | Predicted E3 ubiquitin ligase |
| 2674758 | -22.74 | | | Gaa1-like, GPI transamidase component | integral to membrane, GPI-anchor transamidase complex | Cellular processes and signaling | Posttranslational modification, protein turnover, chaperones | Glycosylphosphatidylinositol anchor attachment protein GAA1 |
| 2565418 | -21.89 | hydrolase activity | | NUDIX hydrolase, core | | Information storage and processing | Replication, recombination and repair | Peroxisomal NUDIX hydrolase |
| 2269164 | -2.40 | nucleic acid binding | | RNA recognition motif, RNP-1 | | Information storage and processing | RNA processing and modification | Splicing factor RNPS1 |
| 2670584 | -22.33 | endopeptidase Clp activity | proteolysis and peptidolysis | Peptidase S14, ClpP | | Information storage and processing | RNA processing and modification | Splicing coactivator SRm160/300, subunit SRm160 |
| 2600463 | -23.57 | glycolipid 2-alpha-mannosyltransferase activity | protein amino acid glycosylation, protein modification | Glycosyl transferase, family 15 | membrane | Information storage and processing | RNA processing and modification | Splicing coactivator SRm160/300, subunit SRm300 |

| | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| 258538 | -2.21 | FAD binding, NADPH binding, dimethylaniline monooxygenase (N-oxide-forming) activity | | Flavin-containing monooxygenase FMO, Dimethylaniline monooxygenase, N-oxide-forming | microsome | Metabolism | Secondary metabolites biosynthesis, transport and catabolism | Flavin-containing monooxygenase |
| 2602031 | -2.95 | monooxygenase activity, peptidase activity, heme binding, iron ion binding | electron transport, proteolysis and peptidolysis | Cytochrome P450, E-class, group I | | Metabolism | Secondary metabolites biosynthesis, transport and catabolism | Cytochrome P450 CYP2 subfamily |
| 2706126 | -3.02 | catalytic activity, binding | metabolism | NAD(P)-binding | | Metabolism | Secondary metabolites biosynthesis, transport and catabolism | Alcohol dehydrogenase, class V |
| 2680003 | -3.98 | catalytic activity, ammonia ligase activity | biosynthesis, phenylalanine catabolism | L-Aspartase-like, Phenylalanine/ histidine ammonia-lyase | cytoplasm | Metabolism | Secondary metabolites biosynthesis, transport and catabolism | Phenylalanine and histidine ammonia-lyase |
| 2664870 | -4.47 | hydrogen ion transporter activity, O-methyltransferase activity, zinc ion binding, DNA binding, transcription factor activity | regulation of transcription, DNA-dependent, transcription | ATPase, F1/V1/A1 complex, alpha/beta subunit, nucleotide-binding; O-methyltransferase, family 3; Fungal specific transcription factor | proton-transporting two-sector ATPase complex, nucleus | Metabolism | Secondary metabolites biosynthesis, transport and catabolism | O-methyl transferase |
| 2016177 | -21.43 | transporter activity, binding, oxidoreductase activity | | Lipocalin, FAD dependent oxidoreductase | | Metabolism | Secondary metabolites biosynthesis, transport and catabolism | Flavin-containing monooxygenase |
| 2748945 | -22.57 | nucleoside triphosphatase activity, nucleotide binding, ATPase activity, coupled to transmembrane movement of substances | transport | AAA+ ATPase, core; ABC transporter, transmembrane region | integral to membrane | Metabolism | Secondary metabolites biosynthesis, transport and catabolism | Multidrug resistance -associated protein/ mitoxantrone resistance protein, ABC super family |

| | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| 2748917 | -23.08 | nucleoside triphosphatase activity, nucleotide binding, ATPase activity, coupled to transmembrane movement of substance | transport | AAA+ ATPase, core; ABC transporter | | Metabolism | Secondary metabolites biosynthesis, transport and catabolism | Multidrug resistance-associated protein/mitoxantrone resistance protein, ABC super family |
| 2341189 | -24.18 | ammonia ligase activity, ammonia-lyase activity | biosynthesis, phenylalanine catabolism | L-Aspartase-like; Phenylalanine/histidine ammonia-lyase | cytoplasm | Metabolism | Secondary metabolites biosynthesis, transport and catabolism | Phenylalanine and histidine ammonia-lyase |
| 2674266 | -26.47 | | | | | Metabolism | Secondary metabolites biosynthesis, transport and catabolism | Flavin-containing monooxygenase |
| 2442962 | -2.01 | protein kinase activity, protein serine/threonine kinase activity, ATP binding | protein amino acid phosphorylation | Protein kinase, core, Serine/threonine protein kinase-related | | Cellular processes and signaling | Signal transduction mechanisms | Serine/threonine protein kinase |
| 2749526 | -23.25 | | | | | Cellular processes and signaling | Signal transduction mechanisms | Predicted calmodulin-binding protein |
| 2347021 | -23.50 | | autophagy | Autophagy protein 6 | | Cellular processes and signaling | Signal transduction mechanisms | Beclin-like protein |
| 2669964 | -23.90 | adenosylmethionine decarboxylase activity | spermine biosynthesis, spermidine biosynthesis | S-adenosylmethionine decarboxylase | | Cellular processes and signaling | Signal transduction mechanisms | S-adenosylmethionine decarboxylase |
| 2695958 | -21.11 | | | | | Information storage and processing | Transcription | RNA polymerase III transcription factor TFIIIC |
| 2707183 | -24.28 | FAD binding, rhodopsin-like receptor activity, binding, transporter activity, oxidoreductase activity | electron transport, signal transduction, G-protein coupled receptor protein signaling pathway, transport | Pyridine nucleotide-disulphide oxidoreductase; GPCR, rhodopsin-like | integral to membrane | Metabolism | Transcription | Nuclear receptor coregulator SMRT/SMRTER, contains Myb-like domains |
| 2340137 | -2.01 | | | | | | | |

| | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| 2599836 | -2.04 | | | | | | | |
| 2468516 | -2.16 | | | | | | | |
| 2679050 | -2.16 | metal ion transporter activity | metal ion transport | Mg$^{2+}$ transporter protein, CorA-like | membrane | | | |
| 2231504 | -2.16 | | | | | | | |
| 2598923 | -2.41 | catalytic activity, hydrolase activity, acting on carbon-nitrogen (but not peptide) bonds | carbohydrate metabolism | Glycoside hydrolase/ deacetylase, beta/alpha-barrel | | | | |
| 2500013 | -2.43 | transition metal ion binding, motor activity, motor activity, heme binding, ATP binding, transferase activity | | Cytochrome b5, Myosin head, motor region | myosin | | | |
| 2602828 | -2.43 | catalytic activity, hydrolase activity, acting on carbon-nitrogen (but not peptide) bonds | carbohydrate metabolism | Glycoside hydrolase/ deacetylase, beta/alpha-barrel | | | | |
| 2474382 | -2.45 | ferrous iron binding, oxidoreductase activity | electron transport, aromatic compound metabolism | Extradiol ring-cleavage dioxygenase | | | | |
| 2698202 | -2.57 | | | | | | | |
| 2564910 | -2.59 | | | | integral to membrane, membrane | | | Protein of unknown function DUF846, eukaryotic |
| 2565384 | -2.68 | catalytic activity, binding, oxidoreductase activity | metabolism | NAD(P)-binding | | | | |
| 2597807 | -2.91 | ubiquitin-like-protein ligase activity | | Ubiquitin-conjugating enzyme, E2 | | | | |

| | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| 2484129 | -2.92 | cAMP-specific phosphodiesterase activity, 3',5'-cyclic-nucleotide phosphodiesterase activity | cAMP catabolism | Cyclic-AMP phosphodiesterase, class-II | | | | |
| 2487587 | -2.96 | | | | | | | |
| 2597346 | -3.09 | | | | | | | |
| 2693614 | -3.28 | catalytic activity, protein binding, pyridoxal phosphate binding | protein complex assembly, transport, metabolism, intracellular protein transport, vesicle-mediated transport | Pyridoxal phosphate-dependent enzyme, beta subunit, Clathrin adaptor | membrane coat | | | |
| 2742390 | -3.73 | catalytic activity | | Six-hairpin glycosidase-like | | | | |
| 2676304 | -3.75 | protein binding, zinc ion binding | | Zinc finger, PHD-type | | | | |
| 2746425 | -4.10 | | | | | | | |
| 2441717 | -20.61 | protein modification | | Ubiquitin | | | | |
| 2590616 | -20.97 | | | | | | | |
| 2676307 | -21.21 | | | | | | | |
| 2503274 | -21.21 | | | | | | | |
| 2539960 | -21.50 | hydrolase activity, hydrolyzing O-glycosyl compounds | carbohydrate metabolism | Glycoside hydrolase, family 3 | | | | |
| 2673553 | -21.94 | catalytic activity, hydrolase activity, acting on carbon-nitrogen (but not peptide) bonds | carbohydrate metabolism | Glycoside hydrolase/deacetylase, beta/alpha-barrel | | | | |
| 2601363 | -22.19 | metalloendopeptidase activity | proteolysis and peptidolysis | Peptidase M35, deuterolysin | | | | |
| 2622441 | -22.31 | metal ion transporter activity | metal ion transport | Mg$^{2+}$ transporter protein, CorA-like | membrane | | | |
| 2677817 | -22.39 | | | | | | | |
| 2603372 | -22.56 | | | | | | | |

| | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| 2744761 | -22.64 | DNA binding, transcription factor activity, zinc ion binding | Transcription, regulation of transcription, DNA-dependent | Fungal specific transcription factor | nucleus | | | |
| 2558376 | -22.67 | catalytic activity, binding | metabolism | NAD(P)-binding | | | | |
| 2600095 | -22.93 | | | | | | | |
| 2467057 | -22.94 | | | | | | | |
| 2688245 | -23.24 | protein binding, zinc ion binding | peroxisome organization and biogenesis | Zinc finger, RING-type; Pex, | peroxisomal membrane | | | |
| 2337178 | -23.31 | | | | | | | |
| 2197031 | -23.37 | | | | | | | |
| 2519099 | -23.53 | heme binding | | Indoleamine 2,3-dioxygenase | | | | |
| 2605007 | -23.57 | | | | | | | |
| 2186683 | -23.59 | | | | | | | |
| 2631069 | -23.70 | | | | | | | |
| 2591615 | -23.84 | | | | | | | |
| 2676799 | -23.87 | | | | | | | |
| 2450756 | -23.98 | catalytic activity, binding | metabolism | NAD(P)-binding | | | | |
| 1181308 | -24.01 | | | | | | | |
| 2698599 | -24.02 | | | | | | | |
| 2583437 | -24.71 | oxidoreductase activity | | FAD dependent oxidoreductase | | | | |
| 2706252 | -24.72 | transporter activity | oligopeptide transport | TGF-beta receptor, type I/II extracellular region | membrane | | | |
| 2596228 | -24.80 | | | | | | | |
| 2439672 | -24.90 | | | | | | | |

## Table S8. Number of proteins regulated in dikaryon vs. monokaryon of *S. commune*

| **KOG Group*** | **Dikaryon vs. monokaryon 12-43** | | **Dikaryon vs. monokaryon 4-39** | |
|---|---|---|---|---|
| | **High abundance proteins** | **Low abundance proteins** | **High abundance proteins** | **Low abundance proteins** |
| **Cellular processes and signaling** | 5 | 15 | 2 | 8 |
| **Information storage and processing** | 6 | 8 | 3 | 13 |
| **Metabolism** | 15 | 24 | 5 | 22 |
| **Poorly characterized** | 2 | 10 | 5 | 9 |
| **Not available** | 15 | 40 | 11 | 42 |
| **Total** | 43 | 97 | 26 | 94 |

## Table S9. Number of proteins regulated in primordia vs. monokaryon of *S. commune*

| KOG Group* | Primordia vs. monokaryon 12-43 | | Primordia vs. monokaryon 4-39 | |
|---|---|---|---|---|
| | High abundance proteins | Low abundance proteins | High abundance proteins | Low abundance proteins |
| **Cellular processes and signaling** | 14 | 27 | 23 | 30 |
| **Information storage and processing** | 8 | 13 | 18 | 26 |
| **Metabolism** | 36 | 36 | 58 | 65 |
| **Poorly characterized** | 10 | 15 | 25 | 27 |
| **Not available** | 56 | 72 | 69 | 82 |
| **Total** | 124 | 163 | 193 | 230 |

## Table S10. Number of proteins regulated in fruiting body vs. monokaryon of *S. commune*

| KOG Group* | Fruiting body vs. monokaryon 12-43 | | Fruiting body vs. monokaryon 4-39 | |
|---|---|---|---|---|
| | High abundance proteins | Low abundance proteins | High abundance proteins | Low abundance proteins |
| **Cellular processes and signaling** | 15 | 107 | 25 | 118 |
| **Information storage and processing** | 14 | 49 | 17 | 75 |
| **Metabolism** | 33 | 145 | 48 | 170 |
| **Poorly characterized** | 18 | 50 | 27 | 53 |
| **Not available** | 81 | 132 | 98 | 167 |
| **Total** | 161 | 483 | 215 | 583 |

* KOG Group was analyzed using the JGO schco1

## Table S11. Induced proteins in dikaryon vs. monokaryon 12-43

| Schco3 ID | Fold change | Go Desc molecular function | Go Desc Biological Process | Interpro Desc | KOG Group |
|---|---|---|---|---|---|
| 2437587 | 3.963 | phosphoribosylanthranilate isomerase activity, catalytic activity, anthranilate synthase activity, indole-3-glycerol-phosphate synthase activity | glutamine metabolism, biosynthesis, tryptophan metabolism, metabolism | N-(5'phosphoribosyl) anthranilate isomerase (PRAI); Glutamine amidotransferase superfamily; Anthranilate synthase component II/delta crystallin; Ribulose-phosphate binding barrel; Indole-3-glycerol phosphate synthase, central region | |
| 2612825 | 2.975 | catalytic activity, branched-chain-amino-acid transaminase activity | branched chain family amino acid metabolism, metabolism | Aminotransferase, class IV; Branched-chain amino acid aminotransferase II | |
| 2681708 | 2.015 | catalytic activity, 3-phosphoshikimate 1-carboxyvinyltransferase activity, hydroxymethylglutaryl-CoA reductase (NADPH) activity, shikimate 5-dehydrogenase activity, shikimate kinase activity binding, ATP binding | coenzyme A metabolism, aromatic amino acid family biosynthesis, metabolism | RNA 3'-terminal phosphate cyclase/enolpyruvate transferase, alpha/beta; Quinate/shikimate 5-dehydrogenase/glutamyl-tRNA reductase; Shikimate-5-dehydrogenase | |
| 2472473 | 4.344 | transferase activity, transferring alkyl or aryl | amino acid metabolism | Cys/Met metabolism, pyridoxal phosphate-dependent enzyme; | |

| | | | | | |
|---|---|---|---|---|---|
| | | (other than methyl) groups, pyridoxal phosphate binding | | O-acetylhomoserine/O-acetylserine sulfhydrylase | |
| 2118192 | 3.947 | chitinase activity, hydrolase activity, hydrolyzing O-glycosyl compounds | chitin catabolism, carbohydrate metabolism | Chitinase II; Glycoside hydrolase, family 18, catalytic domain | |
| 2600607 | 3.157 | | | | |
| 2688579 | 2.384 | | | | |
| 2679442 | 2.091 | catalytic activity | | Six-hairpin glycosidase-like | |
| 2502077 | 2.363 | | | | |
| 2595827 | 2.675 | catalytic activity, binding, oxidoreductase activity | metabolism | Aldehyde dehydrogenase; NAD(P)-binding | |
| 2637911 | 2.044 | aldehyde dehydrogenase [NAD(P)] activity, oxidoreductase activity | aldehyde metabolism, metabolism | Aldehyde dehydrogenase; Aldehyde/histidinol dehydrogenase | |
| 2640974 | 4.681 | | | | |
| 2660823 | 6.074 | metalloendopeptidase activity | proteolysis and peptidolysis | Peptidase M35, deuterolysin | |
| 2587232 | 2.182 | sugar porter activity, transporter activity | carbohydrate transport, transport | Major facilitator superfamily; Sugar transporter; General substrate transporter | |
| 2600367 | 2.657 | FAD binding, iron ion binding, oxidoreductase activity | electron transport | Ferric reductase-like transmembrane component, N-terminal | |
| 2721824 | 2.524 | protein binding | cell wall organization and biogenesis, protein localization | Vacuolar protein sorting-associated protein | Cellular process and signaling |
| 2197094 | 3.152 | catalytic activity | metabolism | AMP-dependent synthetase and ligase | |
| 2679197 | 2.630 | hydrolase activity, histone deacetylase activity | histone deacetylation | Histone deacetylase; Metallophosphoesterase | |
| 2677767 | 2.053 | nicotinate-nucleotide diphosphorylase (carboxylating) activity | nicotinamide adenine dinucleotide biosynthesis, pyridine nucleotide biosynthesis | Quinolinate phosphoribosyl transferase; Nicotinate-nucleotide pyrophosphorylase | |
| 2603157 | 4.166 | aspartic-type endopeptidase activity, transporter activity, pepsin A activity | oligopeptide transport, proteolysis and peptidolysis | Peptidase A1; Peptidase aspartic, active site; TGF-beta receptor, type I/II extracellular region | |
| 2600956 | 3.252 | protein binding, FAD binding, oxidoreductase activity, acting on CH-OH group of donors, binding, ATP binding, nucleotide binding, nucleoside triphosphatase activity | peroxisome organization and biogenesis | Peroxisome biogenesis factor 1; Glucose-methanol-choline oxidoreductase; Aspartate decarboxylase-like fold; AAA+ ATPase | |
| 2677822 | 3.249 | ligase activity, ATP binding | mismatch repair | Carboxyl transferase; Acetyl-coenzyme A carboxyltransferase; DNA mismatch repair protein | |
| 2675600 | 4.389 | hydrolase activity | | NUDIX hydrolase, core | |
| 2678866 | 2.058 | | | | |
| 2599439 | 2.337 | monooxygenase activity, NADH dehydrogenase (ubiquinone) activity, heme binding, iron ion binding, electron carrier activity | electron transport | Cytochrome P450 | |

| | | | | | |
|---|---|---|---|---|---|
| 2505976 | 2.476 | DNA binding, transcription regulator activity, DNA-directed RNA polymerase activity, nucleic acid binding, zinc ion binding | transcription, regulation of transcription | DNA-directed RNA polymerase, M/15 kDa subunit; Zinc finger, TFIIS-type | |
| 2540534 | 2.977 | | | | |
| 2354737 | 2.855 | transferase activity | | Trimeric LpxA-like | |
| 2676857 | 6.491 | catalytic activity | | Six-hairpin glycosidase-like | |
| 2625719 | 5.036 | aminopeptidase activity, prolyl aminopeptidase activity | proteolysis and peptidolysis | Peptidase S33, prolyl aminopeptidase | |
| 2476158 | 4.947 | binding | | C-type lectin | |
| 2748821 | 4.819 | catalytic activity, hydrolase activity, acting on carbon-nitrogen (but not peptide) bonds | carbohydrate metabolism | Polysaccharide deacetylase; Glycoside hydrolase/deacetylase, beta/alpha-barrel | |
| 2602331 | 4.687 | | | | |
| 2154417 | 4.254 | | | | |
| 2684270 | 3.368 | | | | |
| 2179879 | 3.206 | copper ion binding, oxidoreductase activity | | Multicopper oxidase, copper-binding site | |
| 2626480 | 3.059 | | | | |
| 2581083 | 2.754 | RNA binding, nucleic acid binding | mRNA metabolism | Polyadenylate binding protein, human types 1, 2, 3, 4; RNA recognition motif, RNP-1 | |
| 1105701 | 2.722 | | | | |
| 2602828 | 2.505 | catalytic activity, hydrolase activity, acting on carbon-nitrogen (but not peptide) bonds | carbohydrate metabolism | Polysaccharide deacetylase; Glycoside hydrolase/deacetylase, beta/alpha-barrel | |
| 2450251 | 2.405 | binding | transport | Mitochondrial substrate carrier | |
| 2705026 | 2.217 | catalytic activity, glyceraldehyde-3-phosphate dehydrogenase (phosphorylating) activity, binding, glyceraldehyde-3-phosphate dehydrogenase activity | glycolysis, glucose metabolism, metabolism | Glyceraldehyde-3-phosphate dehydrogenase, type I; NAD(P)-binding | |
| 2529447 | 2.016 | hydrolase activity, hydrolyzing O-glycosyl compounds | carbohydrate metabolism | Glycoside hydrolase, family 5 | |

## Table S12. Induced proteins in dikaryon vs. monokaryon 4-39

| Schco3 ID | Fold change | Go Desc molecular function | Go Desc Biological Process | Interpro Desc | KOG Group |
|---|---|---|---|---|---|
| 2472473 | 5.064 | transferase activity, transferring alkyl or aryl (other than methyl) groups, pyridoxal phosphate binding | amino acid metabolism | O-acetylhomoserine/O-acetylserine sulfhydrylase; Cys/Met metabolism, pyridoxal phosphate-dependent enzyme | |
| 2706358 | 2.153 | NAD ADP-ribosyltransferase activity, mevalonate kinase activity, phosphotransferase activity, alcohol group as acceptor, ATP binding, pyridoxal phosphate binding, kinase activity | amino acid metabolism, isoprenoid biosynthesis metabolism | Poly (ADP-ribose) polymerase, catalytic region; Mevalonate kinase; Mevalonate and galactokinase; GHMP kinase; GHMP kinase, ATP-binding, conserved site; Cys/Met metabolism, pyridoxal phosphate-dependent enzyme; | |
| 2701913 | 2.443 | | | | |
| 2660823 | 4.722 | metalloendopeptidase activity | proteolysis and peptidolysis | Peptidase M35, deuterolysin | |
| 2506333 | 4.568 | cytochrome-c oxidase activity | electron transport | Cytochrome c oxidase, subunit Vib | |

| | | | | | |
|---|---|---|---|---|---|
| 2680499 | 3.906 | cytochrome-c oxidase activity | electron transport | Cytochrome c oxidase, subunit VIb | |
| 2680216 | 3.264 | nucleic acid binding, zinc ion binding | | Zinc finger, C2H2-type; Zinc finger, C2H2-like | |
| 2678477 | 2.042 | monooxygenase activity, iron ion binding, sugar porter activity, caspase activity, heme binding, transporter activity, hydrolase activity, oxidoreductase activity | electron transport, carbohydrate transport, proteolysis and peptidolysis, transport | Cytochrome P450; Sugar transporter; Peptidase C14, caspase catalytic; General substrate transporter; Major facilitator superfamily, Sugar transporter, conserved site, Metallophosphoesterase; Peptidase C14, caspase catalytic | |
| 2681970 | 3.817 | catalytic activity | metabolism | AMP-dependent synthetase and ligase | |
| 2600532 | 2.230 | DNA binding, transporter activity, hydrolase activity, acting on carbon-nitrogen (but not peptide) bonds, RNA polymerase II transcription factor activity | transport, regulation of transcription | Histone-fold, Major facilitator superfamily, Metal-dependent hydrolase, composite, TAFII28-like protein, TAFII28-like protein | |
| 2599287 | 2.316 | peptidase activity | proteolysis and peptidolysis | Peptidase M28 | |
| 2699460 | 3.773 | helicase activity, RNA binding, nucleic acid binding, ribonuclease III activity, ATP dependent helicase activity, ATP binding | RNA processing | DNA/RNA helicase, C-terminal; Ribonuclease III; DNA/RNA helicase, DEAD/DEAH box type, N-terminal; DNA/RNA helicase, DEAD/DEAH box type, N-terminal | |
| 2603198 | 6.849 | | | | |
| 2528965 | 3.470 | double-stranded RNA binding, RNA binding, ribonuclease III activity | RNA processing, intracellular | Double-stranded RNA binding; Ribonuclease III | |
| 2750512 | 3.159 | nucleic acid binding | | Polynucleotidyl transferase, Ribonuclease H fold | |
| 2579584 | 4.621 | subtilase activity, serine-type peptidase activity | proteolysis and peptidolysis | Peptidase S8 and S53; subtilisin; kexin; sedolisin; propeptide | |
| 2676242 | 3.614 | DNA binding, NAD-dependent histone deacetylase activity, zinc ion binding | chromatin silencing, regulation of transcription, DNA-dependent, protein amino acid deacetylation | NAD-dependent histone deacetylase, silent information regulator Sir2 | |
| 2154417 | 3.593 | | | | |
| 2602828 | 3.574 | catalytic activity, hydrolase activity, acting on carbon-nitrogen (but not peptide) bonds | carbohydrate metabolism | Glycoside hydrolase/deacetylase, beta/alpha-barrel; Polysaccharide deacetylase | |
| 2626231 | 3.203 | catalytic activity, transcriptional repressor activity binding | regulation of nitrogen utilization metabolism | NAD(P)-binding; NmrA-like | |
| 2753486 | 3.188 | | | | |
| 2179879 | 3.055 | copper ion binding, oxidoreductase activity | | Multicopper oxidase, type 2; Multicopper oxidase, type 3; Multicopper oxidase, copper-binding site; Multicopper oxidase, type 1 | |
| 2684270 | 2.991 | | | | |

| | | | | | |
|---|---|---|---|---|---|
| 2625719 | 2.887 | aminopeptidase activity, prolyl aminopeptidase activity, | proteolysis and peptidolysis | Peptidase S33, prolyl aminopeptidase; Peptidase S33, tricorn interacting factor 1 | |
| 2680434 | 2.187 | hydrolase activity, hydrolyzing O-glycosyl compounds | carbohydrate metabolism | Glycoside hydrolase, family 5 | |
| 2704482 | 2.060 | | | | |

**Table S13. Repressed proteins in dikaryon vs. monokaryon 12-43**

| Schco3 ID | Fold change | Go Desc molecular function | Go Desc Biological Process | Interpro Desc | KOG Group |
|---|---|---|---|---|---|
| 2725707 | -2.041 | protein kinase activity, protein-tyrosine kinase activity, protein serine/threonine kinase activity, ATP binding | protein amino acid phosphorylation | Protein kinase; Serine/threonine protein kinase-related; Protein kinase ATP binding; Tyrosine protein kinase | |
| 2603198 | -2.214 | | | | |
| 2249278 | -2.222 | | | | |
| 2680703 | -2.266 | glucosylceramidase activity protein binding, binding | lysosome organization and biogenesis, protein complex assembly, sphingolipid metabolism, intracellular protein transport, vesicle-mediated transport | Glycoside hydrolase, family 30; Clathrin/coatomer adaptor, adaptin-like; Clathrin/coatomer adaptor, adaptin-like; | |
| 2354629 | -2.270 | GTP binding | protein transport, small GTPase mediated signal transduction, intracellular protein transport | ARF/SAR superfamily; Ras GTPase; ADP-ribosylation factor; Ras small GTPase, Rab type | Cellular processes and signaling |
| 2675738 | -2.320 | intracellular protein transport | | NSF attachment protein | |
| 1141646 | -2.352 | GTPase activity, structural molecule activity, GTP binding | microtubule-based movement, microtubule-based process | Tubulin/FtsZ; Alpha tubulin | |
| 2674968 | -2.364 | nucleic acid binding, ATP binding, zinc ion binding | | Dephospho-CoA kinase; Zinc finger, C2H2-type | |
| 2640508 | -2.450 | phosphatidylinositol phosphate kinase activity | phosphatidylinositol metabolism | Phosphatidylinositol-4-phosphate 5-kinase, core | |
| 2572877 | -2.693 | peptidase activity | proteolysis and peptidolysis | Peptidase M28 | |
| 2674326 | -2.721 | microtubule motor activity, ATP binding | microtubule-based movement | Kinesin, motor region | |
| 2596185 | -2.728 | protein binding, GTP binding, GTPase activity | nucleocytoplasmic transport, signal transduction, small GTPase mediated signal transduction, protein transport | Ran GTPase; Small GTP-binding protein; Ras small GTPase, Ras type; Ras small GTPase, Rho type; Ras small GTPase, Rab type | |

| | | | | | |
|---|---|---|---|---|---|
| 2678401 | -3.302 | GTPase activity, structural molecule activity, GTP binding | microtubule-based movement, microtubule-based process | Tubulin/FtsZ; Beta tubulin | |
| 2127007 | -3.441 | metal ion binding | | Class II aldolase/adducin | |
| 2702602 | -3.938 | calcium ion binding | | Calcium-binding EF-hand | |
| 2680463 | -2.152 | helicase activity, catalytic activity, CTP synthase activity, nucleic acid binding, ATP dependent helicase activity, ATP binding | pyrimidine nucleotide biosynthesis | DNA/RNA helicase; Glutamine amidotransferase class-I, CTP synthase | |
| 2743884 | -2.563 | protein kinase activity, transition metal ion binding, protein-tyrosine kinase activity, protein serine/threonine kinase activity, ATP binding, oxidoreductase activity | protein amino acid phosphorylation | Protein kinase; Serine/threonine protein kinase-related; Protein kinase ATP binding, conserved site; Ferritin/ribonucleotide reductase-like; Ferritin/ribonucleotide reductase-like | |
| 2680491 | -2.833 | protein binding, ATPase activity, ATP binding | DNA metabolism | ABC transporter-like; RecF/RecN/SMC protein; SMCs flexible hinge | |
| 2491982 | -2.887 | structural constituent of ribosome, RNA binding | protein biosynthesis | Ribosomal protein L23, conserved site | |
| 2601895 | -2.961 | calcium ion binding, ATP binding | mismatch repair | DNA mismatch repair protein; ATP-binding region, ATPase-like; Calcium-binding EF-hand | |
| 2488899 | -2.977 | structural constituent of ribosome | protein biosynthesis | Ribosomal protein S27e | |
| 2663340 | -3.415 | structural constituent of ribosome | protein biosynthesis | Ribosomal protein S16 | |
| 2195418 | -4.194 | | | | |
| 2502355 | -2.021 | acyl-CoA binding | | Acyl-CoA-binding protein, ACBP | |
| 2680115 | -2.078 | catalytic activity | | UBA/THIF-type NAD/FAD binding fold | |
| 2131899 | -2.134 | ATP binding | anion transport | Anion-transporting ATPase | |
| 2596034 | -2.144 | regulation of signal transduction, response to biotic stimulus | | TAP42-like protein | |
| 2255769 | -2.203 | | | Translationally controlled tumour-associated TCTP | |
| 2436819 | -2.251 | chromatin binding, binding, ATP binding, nucleotide binding, nucleoside triphosphatase activity, ATPase activity | chromatin assembly/disassembly | Chromo domain; Armadillo-type fold; ABC transporter-like; AAA+ ATPase, core | |
| 2597073 | -2.276 | amino acid transport, transport | | Amino acid permease | |
| 2700638 | -2.282 | purine-nucleoside phosphorylase activity, transferase activity, transferring pentosyl groups | nucleobase, nucleoside, nucleotide and nucleic acid metabolism | Inosine guanosine and xanthosine phosphorylase; Purine phosphorylase, family 2 | |
| 2535945 | -2.289 | phosphoribosylaminoimidazolesuccinocarboxamide synthase activity | purine nucleotide biosynthesis | SAICAR synthetase | |
| 2312113 | -2.298 | catalytic activity, binding, coenzyme binding | metabolism | | |
| 2675904 | -2.320 | hydrolase activity, hydrolyzing O-glycosyl compounds, oxidoreductase activity | carbohydrate metabolism | Glycoside hydrolase family 2, immunoglobulin-like beta-sandwich; Aldo/keto reductase | |

| | | | | | |
|---|---|---|---|---|---|
| 2624619 | -2.377 | D-amino-acid oxidase activity, oxidoreductase activity | | D-amino acid oxidase | |
| 2597083 | -2.547 | acyltransferase activity | metabolism | Phospholipid/glycerol acyltransferase | |
| 2674964 | -2.571 | catalytic activity, protein kinase activity, protein serine/threonine kinase activity, ATP binding | protein amino acid phosphorylation, metabolism | Fumarylacetoacetase; Protein kinase; Serine/threonine protein kinase-related | |
| 2551610 | -2.589 | peroxidase activity, ATP binding, nucleoside triphosphatase activity, nucleotide binding, heme binding, ATPase activity, ATPase activity, coupled to transmembrane movement of substances | electron transport, transport, response to oxidative stress | ABC transporter, transmembrane region; AAA+ ATPase, core | |
| 2516399 | -2.782 | calcium ion binding, mannosyl-oligosaccharide 1,2-alpha-mannosidase activity | | Glycoside hydrolase, family 47 | |
| 2676329 | -3.018 | catalytic activity, phospho-N-acetylmuramoyl-pentapeptide-transferase activity, isocitrate lyase activity | carboxylic acid metabolism, metabolism | Isocitrate lyase and phosphorylmutase; Pyruvate/Phosphoenolpyruvate kinase, catalytic core; Glycosyl transferase, family 4 | |
| 2676596 | -3.090 | catalytic activity, carboxyl- and carbamoyltransferase activity, monooxygenase activity, binding, amino acid binding, oxidoreductase activity | electron transport, amino acid metabolism, aromatic compound metabolism, metabolism | Aspartate/ ornithine carbamoyltransferase; Monooxygenase, FAD-binding; Glucose/ribitol dehydrogenase; Pyridine nucleotide-disulphide oxidoreductase, class I | |
| 2695732 | -3.109 | transferase activity, transferring nitrogenous groups, pyridoxal phosphate binding | Aminotransferase, class I and II | | |
| 2680902 | -3.131 | carboxy-lyase activity, pyridoxal phosphate binding | carboxylic acid metabolism | Pyridoxal phosphate-dependent decarboxylase | |
| 2469166 | -3.276 | | | | Information storage and processing |
| 2725570 | -4.260 | catalytic activity | acetyl-CoA metabolism | Acetyl-CoA hydrolase/transferase | |
| 1152047 | -4.326 | catalytic activity | | Epoxide hydrolase-like | |
| 2714417 | -7.186 | | | | |
| 2643023 | -2.122 | FAD binding, oxidoreductase activity, acting on CH-OH group of donors | alcohol metabolism | Glucose-methanol-choline oxidoreductase | |
| 2680350 | -2.165 | transporter activity | transport | Major facilitator superfamily | |
| 2602357 | -2.178 | protein binding | amino acid transport, transport | Amino acid permease, conserved site; BTB/POZ fold | |
| 2595487 | -2.269 | nucleotide binding | tRNA-binding arm | | |
| 2688236 | -2.361 | catalytic activity, binding, ATP binding, nucleotide binding, tRNA ligase activity, oxidoreductase activity | amino acid activation, protein biosynthesis, metabolism | Short-chain dehydrogenase/reductase SDR; Glucose/ribitol dehydrogenase; Aminoacyl-tRNA synthetase, class I, conserved site | |
| 2693747 | -2.572 | transporter activity, binding | signal transduction, transport | Rho GTPase activation protein; Lipocalin | |

| | | | | | |
|---|---|---|---|---|---|
| 2599506 | -2.703 | C-8 sterol isomerase activity | ergosterol biosynthesis | ERG2 and sigma1 receptor-like | |
| 2231044 | -4.393 | | | | |
| 2749685 | -4.803 | sugar porter activity, transporter activity | carbohydrate transport, transport | Sugar transporter; General substrate transporter; Major facilitator superfamily | |
| 2696773 | -5.287 | | | | |
| 2681191 | -2.044 | catalytic activity, hydrolase activity, acting on carbon-nitrogen (but not peptide) bonds, hydrolase activity | metabolism | Haloacid dehalogenase-like hydrolase; Metal-dependent hydrolase, composite; Amidohydrolase 1 | |
| 2702456 | -2.110 | protein kinase activity, protein-tyrosine kinase activity, protein serine/threonine kinase activity, ATP binding | protein amino acid phosphorylation | Protein kinase; Serine/threonine protein kinase-related; Tyrosine protein kinase; Serine/threonine protein kinase | |
| 2597062 | -2.185 | mannose-1-phosphate guanylyltransferase (GDP) activity, nucleotidyltransferase activity | polysaccharide metabolism, biosynthesis | Mannose-6-phosphate isomerase, type II | |
| 2678282 | -2.223 | two-component sensor molecule activity, G-protein coupled photoreceptor activity, protein-histidine kinase activity, two-component response regulator activity, signal transducer activity, ATP binding, transferase activity, transferring phosphorus-containing groups | regulation of transcription, peptidyl-histidine phosphorylation, sensory perception, protein-chromophore linkage, signal transduction, phosphorylation, two-component signal transduction system (phosphorelay), regulation of transcription, DNA-dependent | Signal transduction histidine kinase; Phytochrome; PAS fold-2 | |
| 2608339 | -2.274 | | | | |
| 2678455 | -2.294 | | | | |
| 2674721 | -2.324 | calcium binding | | Calcium-binding EF-hand | |
| 2679225 | -2.366 | DNA binding, transcription factor activity, zinc ion binding | transcription, regulation of transcription, DNA-dependent | Fungal specific transcription factor | |
| 2293651 | -2.382 | | | | |
| 2619664 | -2.485 | | | | |
| 2519451 | -2.522 | | | | |
| 2600031 | -2.599 | hydrolase activity, hydrolyzing O-glycosyl compounds | carbohydrate metabolism | Glycoside hydrolase, family 5 | |
| 2592701 | -2.659 | | | | |
| 2463002 | -2.685 | | | | |
| 2556613 | -2.707 | | | | |
| 2075664 | -2.721 | response to pH, proteolysis and peptidolysis | | Signal transduction response regulator, pH-responsive, PalI/Rim9 | |
| 2540154 | -2.729 | | | | |
| 2574569 | -2.788 | | | | |

| | | | | | |
|---|---|---|---|---|---|
| 2471655 | -2.845 | | | | |
| 2708788 | -2.888 | protein kinase activity, protein-tyrosine kinase activity, protein serine/threonine kinase activity, ATP binding | protein amino acid phosphorylation | Protein kinase; Serine/threonine protein kinase-related; Protein kinase ATP binding, conserved site | |
| 2689252 | -2.930 | nucleotide binding, tRNA ligase activity, ATP binding, oxidoreductase activity | amino acid activation, protein biosynthesis | Aminoacyl-tRNA synthetase, class I, conserved site; FAD dependent oxidoreductase | |
| 2184213 | -2.960 | | | | |
| 2532725 | -2.960 | oxidoreductase activity, acting on paired donors, with oxidation of a pair of donors resulting in the reduction of molecular oxygen to two molecules of water, oxidoreductase activity | lipid metabolism | Fatty acid desaturase | |
| 2546907 | -2.997 | carbohydrate binding, catalytic activity, glucan 1,4-alpha-glucosidase activity | polysaccharide catabolism, polysaccharide metabolism, carbohydrate metabolism | Carbohydrate-binding-like fold; Glycoside hydrolase, carbohydrate-binding; Six-hairpin glycosidase-like | |
| 2528625 | -3.094 | | | | |
| 2594802 | -3.144 | | | | |
| 2472024 | -3.149 | response to pH, proteolysis and peptidolysis | | Signal transduction response regulator, pH-responsive, PalI/Rim9 | |
| 2567631 | -3.154 | | | | |
| 2664870 | -3.383 | hydrogen ion transporter activity, O-methyltransferase activity, zinc ion binding, DNA binding, transcription factor activity | regulation of transcription, DNA-dependent, transcription | O-methyltransferase, family 3; Fungal specific transcription factor | Metabolism |
| 2537084 | -3.481 | | | | |
| 2602110 | -3.573 | | | | |
| 2677399 | -3.737 | catalytic activity, hydrolase activity, acting on ester bonds | metabolism | Alkaline-phosphatase-like, core domain; Phosphoesterase | |
| 2198994 | -3.858 | | | | |
| 2552020 | -4.284 | | | | |
| 2746951 | -4.285 | | | | |
| 2460130 | -4.294 | | | | |
| 2680457 | -4.927 | zinc ion binding | | Zinc finger, MYND-type | |
| 1138359 | -5.483 | | | | |
| 2599368 | -5.487 | | | Peptidase S54, rhomboid | |
| 2742326 | -6.873 | | | | |

## Table S14. Repressed proteins in dikaryon vs. monokaryon 4-39

| Schco3 ID | Fold change | Go Desc molecular function | Go Desc Biological Process | Interpro Desc | KOG Group |
|---|---|---|---|---|---|
| 2680097 | -2.019 | acid phosphatase activity, zinc ion binding, transcription factor activity, ARF guanyl-nucleotide exchange factor activity | regulation of transcription, DNA-dependent | Histidine acid phosphatase; Zinc finger, GATA-type; SEC7-like; | |

| | | | | | |
|---|---|---|---|---|---|
| 2602003 | -2.375 | protein binding, binding, ATP binding, RNA ligase (ATP) activity | tRNA splicing | DEATH-like; Armadillo-type fold; tRNA ligase, phosphodiesterase, fungi; tRNA ligase, kinase, fungi | |
| 2675738 | -2.082 | | intracellular protein transport | NSF attachment protein | |
| 2271597 | -2.460 | intracellular transport | | Ran Binding Protein 1 | |
| 2595504 | -2.067 | protein kinase activity, protein serine/threonine kinase activity, ATP binding, protein-tyrosine kinase activity ATPase activity | protein amino acid phosphorylation, anion transport | Protein kinase, core; Serine/threonine protein kinase-related; Tyrosine protein kinase; Serine/threonine protein kinase related; DNA-binding SAP; ABC transporter-like; Porin | |
| 2677294 | -6.320 | catalytic activity, binding, coenzyme binding | metabolism | NAD-dependent epimerase/dehydratase; NAD(P)-binding | |
| 2678444 | -3.774 | protein kinase activity, protein-tyrosine kinase activity, protein serine/threonine kinase activity, ATP binding, N-acetyltransferase activity | protein amino acid phosphorylation, metabolism | Protein kinase, core; Serine/threonine protein kinase-related; Protein kinase ATP binding, conserved site; Tyrosine protein kinase; Serine/threonine protein kinase; GCN5-related N-acetyltransferase | |
| 2249278 | -2.613 | | | | |
| 2749322 | -2.633 | structural constituent of ribosome | protein biosynthesis | Ribosomal protein S17e | |
| 2488899 | -2.704 | structural constituent of ribosome | protein biosynthesis | Ribosomal protein S27e; Ribosomal protein, zinc-binding; Ribosomal protein S27e; | |
| 2557698 | -2.028 | structural constituent of ribosome | protein biosynthesis | Ribosomal protein L21e | |
| 2678650 | -2.182 | structural constituent of ribosome | protein biosynthesis | Ribosomal protein L31e | |
| 2601895 | -2.924 | calcium ion binding, ATP binding | mismatch repair | Calcium-binding EF-hand; DNA mismatch repair protein, C-terminal; ATP-binding region, ATPase-like | |
| 2681298 | -2.060 | thiamin diphosphokinase activity, ATP binding, Nucleotide binding, glycine-tRNA ligase activity, tRNA ligase activity | thiamin diphosphate biosynthesis, amino acid activation, thiamin metabolism, protein biosynthesis | Thiamin pyrophosphokinase, catalytic region; Thiamin pyrophosphokinase, vitamin B1-binding region; Thiamin pyrophosphokinase; Glycyl-tRNA synthetase, alpha2 dimer; Aminoacyl-tRNA synthetase, class II (G, H, P and S); Anticodon-binding; Aminoacyl-tRNA synthetase, class II; | |
| 2696887 | -2.823 | protein binding, DNA binding, transcription factor activity, oxidoreductase activity | electron transport, regulation of transcription, DNA-dependent | WW/Rsp5/WWP; Histone-fold; Bacterial regulatory protein, LuxR; FMN-dependent alpha-hydroxy acid dehydrogenase; | |
| 2623763 | -3.977 | | | SET | Information storage and processing |
| 2620095 | -2.136 | | | | |
| 2746016 | -2.379 | protein binding | | Leucine-rich repeat | |
| 2680491 | -2.252 | protein binding, ATPase activity, ATP binding | DNA metabolism | SMCs flexible hinge; ABC transporter-like; RecF/RecN/SMC protein, N-termina | |
| 2602338 | -2.556 | tRNA binding, tRNA-pseudouridine synthase I activity, nucleic acid binding, ATP binding, DNA binding, ATPase activity | pseudouridine synthesis, tRNA processing | tRNA pseudouridine synthase; DNA/RNA helicase, C-terminal; SNF2-related; ABC transporter-like | |

| | | | | | |
|---|---|---|---|---|---|
| 2106720 | -2.058 | ribosome binding | mature ribosome assembly | Translation initiation factor IF6 | |
| 2455051 | -2.072 | catalytic activity | metabolism | Thiolase-like | |
| 2599143 | -2.061 | catalytic activity | metabolism | Thiolase-like | |
| 2744912 | -3.089 | hydrolase activity, hydrolyzing O-glycosyl compounds | carbohydrate metabolism | Glycoside hydrolase, family 3, N-terminal; Glycoside hydrolase, family 3, C-terminal | |
| 2703762 | -2.180 | protein kinase activity, protein serine/threonine kinase activity, ATP binding | actin filament polymerization, protein amino acid phosphorylation | Protein kinase, core; Serine/threonine protein kinase; Serine/threonine protein kinase; ARP23 complex 20 kDa subunit; Protein kinase, core | |
| 2697037 | -2.312 | catalytic activity, cofactor binding, oxidoreductase activity, acting on the CH-OH group of donors, NAD or NADP as acceptor, binding | metabolism | NAD(P)-binding; D-isomer specific 2-hydroxyacid dehydrogenase, NAD-binding; D-isomer specific 2-hydroxyacid dehydrogenase, catalytic region; | |
| 2737131 | -2.052 | FAD binding, oxidoreductase activity | electron transport | Pyridine nucleotide-disulphide oxidoreductase, NAD-binding region; Pyridine nucleotide-disulphide oxidoreductase, class I | |
| 2666754 | -2.395 | glutamate-ammonia ligase activity | glutamine biosynthesis, nitrogen metabolism | Glutamine synthetase, catalytic region; Glutamine synthetase, beta-Grasp | |
| 2598771 | -2.948 | oxidoreductase activity | electron transport | FMN-dependent alpha-hydroxy acid dehydrogenase; FMN-dependent alpha-hydroxy acid dehydrogenase, active site | |
| 2530747 | -3.669 | | | | |
| 2695732 | -3.087 | transferase activity, transferring nitrogenous groups, pyridoxal phosphate binding | biosynthesis | Aminotransferase, class I and II | |
| 2679035 | -3.347 | protein binding | | BAR | |
| 2676596 | -2.447 | catalytic activity, carboxyl- and carbamoyltransferase activity, monooxygenase activity, binding, oxidoreductase activity | electron transport, amino acid metabolism, aromatic compound metabolism, metabolism | NAD(P)-binding; Aspartate/ornithine carbamoyltransferase; Monooxygenase, FAD-binding; NAD(P)-binding; Glucose/ribitol dehydrogenase; Pyridine nucleotide-disulphide oxidoreductase, class I; Aromatic-ring hydroxylase; Short-chain dehydrogenase/reductase SDR | |
| 2273629 | -2.006 | protein binding | cell communication | Phox-like | |
| 2535945 | -2.682 | phosphoribosylaminoimidazolesuccinocarboxamide synthase activity | purine nucleotide biosynthesis | SAICAR synthetase | |
| 2675904 | -2.393 | hydrolase activity, hydrolyzing O-glycosyl compounds, oxidoreductase activity | carbohydrate metabolism | Glycoside hydrolase family 2, immunoglobulin-like beta-sandwich; Aldo/keto reductase | |
| 2489166 | -2.306 | protein binding, GTP binding | cell cycle | Septin; Cell division/GTP binding protein | |
| 2583245 | -2.377 | serine carboxypeptidase activity, serine-type peptidase activity | proteolysis and peptidolysis | Peptidase S10, serine carboxypeptidase; Peptidase S26A, signal peptidase I | |
| 2595248 | -2.455 | serine carboxypeptidase activity | proteolysis and peptidolysis | Peptidase S10, serine carboxypeptidase | |
| 2707033 | -2.540 | oxidoreductase activity | | Aldo/keto reductase | |
| 2569226 | -3.171 | oxidoreductase activity | | Aldo/keto reductase | |

| | | | | | |
|---|---|---|---|---|---|
| 2546053 | -3.941 | oxidoreductase activity | | Aldo/keto reductase | |
| 2546713 | -3.416 | catalytic activity, binding | metabolism | NAD(P)-binding | Metabolism |
| 2660812 | -3.021 | oxidoreductase activity | negative regulation of transcription | Flavodoxin/nitric oxide synthase; Flavoprotein WrbA | |
| 2549492 | -2.097 | hydrolase activity, hydrolyzing O-glycosyl compounds | carbohydrate metabolism | Glycoside hydrolase, family 5 | |
| 2693747 | -2.942 | transporter activity, binding | transport, signal transduction | Lipocalin; Rho GTPase activation protein | |
| 2480821 | -2.363 | nucleic acid binding | | RNA recognition motif, RNP-1 | |
| 2502834 | -2.277 | hydrolase activity | | Dienelactone hydrolase | |
| 2615761 | -2.673 | | thiamin biosynthesis | Thiamine biosynthesis Thi4 protein | |
| 2688236 | -2.707 | catalytic activity, binding, ATP binding, nucleotide binding, tRNA ligase activity, oxidoreductase activity | amino acid activation, protein biosynthesis, metabolism | NAD(P)-binding; Aminoacyl-tRNA synthetase, class I, conserved site; Short-chain dehydrogenase/reductase SDR; Glucose/ribitol dehydrogenase; | |
| 2599947 | -2.153 | | | | |
| 2573839 | -2.171 | GTPase activity, GTP binding | | Dynamin, GTPase region; Dynamin GTPase effector | |
| 2513636 | -2.013 | | | | |
| 2452507 | -2.017 | | | | |
| 2574569 | -2.023 | | | | |
| 2597062 | -2.044 | mannose-1-phosphate guanylyltransferase (GDP) activity, nucleotidyltransferase activity | polysaccharide metabolism, biosynthesis | Mannose-6-phosphate isomerase, type II, C-terminal; Nucleotidyl transferase | |
| 2598403 | -2.046 | catalytic activity | | Six-hairpin glycosidase-like | |
| 2565384 | -2.092 | catalytic activity, binding, oxidoreductase activity | metabolism | NAD(P)-binding; 6-phosphogluconate dehydrogenase, C-terminal-like | |
| 2701600 | -2.094 | cysteine-type endopeptidase activity, nutrient reservoir activity | proteolysis and peptidolysis | Peptidase, cysteine peptidase active site; Cupin 1 | |
| 2539960 | -2.141 | hydrolase activity, hydrolyzing O-glycosyl compounds | carbohydrate metabolism | Glycoside hydrolase, family 3, N-terminal | |
| 1209921 | -2.172 | | | | |
| 2599368 | -2.210 | | | Peptidase S54, rhomboid | |
| 2672812 | -2.214 | | | | |
| 2508404 | -2.233 | | | | |
| 2692813 | -2.294 | catalytic activity | metabolism | Mandelate racemase/muconate lactonizing enzyme | |
| 2677702 | -2.365 | | | | |
| 2480617 | -2.369 | transcription factor activity, signal transducer activity | signal transduction, regulation of transcription | STAT transcription factor, coiled coil | |
| 2540154 | -2.435 | | | | |
| 2498164 | -2.479 | hydrolase activity | | Alpha/beta hydrolase | |
| 2472024 | -2.488 | | response to pH, proteolysis and peptidolysis | Signal transduction response regulator, pH-responsive, PalI/Rim9 | |
| 2749954 | -2.511 | structural constituent of ribosome | protein biosynthesis, | Ribosomal protein S11 | |
| 1138359 | -2.539 | | | | |
| 2676241 | -2.540 | catalytic activity, transcriptional repressor activity, binding | regulation of nitrogen utilization, metabolism | NAD(P)-binding; NmrA-like | |

| | | | | | |
|---|---|---|---|---|---|
| 2678455 | -2.573 | | | | |
| 2640541 | -2.597 | | | | |
| 2746425 | -2.641 | | | | |
| 2595350 | -2.690 | GTP binding | protein transport, small GTPase mediated signal transduction | Ras GTPase; Ras small GTPase, Ras type; Ras small GTPase, Rho type; Ras small GTPase, Rab type; Small GTP-binding protein; Ras GTPase | |
| 2743762 | -2.790 | | | | |
| 2599803 | -2.821 | glutamate decarboxylase activity, carboxy-lyase activity, transcription factor activity, pyridoxal phosphate binding | carboxylic acid metabolism, glutamate metabolism, regulation of transcription, DNA-dependent | Glutamate decarboxylase; Pyridoxal phosphate-dependent decarboxylase; Homeobox | |
| 2738673 | -2.916 | GTP binding, | protein transport, small GTPase mediated signal transduction, | Ras GTPase; Ras small GTPase Ras type, Ras small GTPase, Rho type; Ras small GTPase, Rab type; Ras GTPase | |
| 2697931 | -2.998 | | | | |
| 2734322 | -3.003 | | | | |
| 2708788 | -3.056 | protein kinase activity, protein-tyrosine kinase activity, protein serine/threonine kinase activity, ATP binding | protein amino acid phosphorylation | Protein kinase, core; Serine/threonine protein kinase-related; Protein kinase ATP binding, conserved site; Tyrosine protein kinase; Serine/threonine protein kinase; Serine/threonine protein kinase, active site | |
| 2740159 | -3.063 | carbohydrate binding, hydrolase activity, hydrolyzing O-glycosyl compounds | carbohydrate metabolism | Carbohydrate-binding family V/XII | |
| 2586789 | -3.323 | | | | |
| 2548110 | -3.494 | | | | |
| 2537084 | -3.625 | | | | |
| 2493604 | -3.697 | | | | |
| 2536636 | -3.826 | | | | |
| 2699394 | -3.925 | | | | |
| 2528310 | -4.217 | | | | |
| 2742326 | -4.354 | | | | |
| 2195418 | -5.045 | | | | |
| 2680457 | -5.084 | zinc ion binding | | Zinc finger, MYND-type | |

### Table S15. Induced proteins in primordia vs. monokaryon 12-43

| **Schco3 ID** | **Fold change** | **Go Desc molecular function** | **Go Desc Biological Process** | **Interpro Desc** | **KOG Group** |
|---|---|---|---|---|---|
| 2749150 | 9.308 | | | Allergen V5/Tpx-1 related | |
| 2660823 | 6.404 | metalloendopeptidase activity | proteolysis and peptidolysis | Peptidase M35, deuterolysin | |
| 2308933 | 5.978 | | | Allergen V5/Tpx-1 related | |
| 2601623 | 4.591 | catalytic activity, carbamoyl-phosphate synthase activity, hydrolase activity | glutamine metabolism, biosynthesis, nitrogen metabolism | Glutamine amidotransferase superfamily; Anthranilate synthase component II/delta crystallin; Carbamoyl phosphate synthase, GATase region | |
| 2110112 | 4.305 | | | Auxin efflux carrier | |

| | | | | | |
|---|---|---|---|---|---|
| 2678477 | 3.200 | monooxygenase activity, iron ion binding, sugar porter activity, caspase activity, transporter activity, heme binding, hydrolase activity, oxidoreductase activity | electron transport, carbohydrate transport, proteolysis and peptidolysis, transport | Cytochrome P450; Sugar transporter; Peptidase C14, caspase catalytic | |
| 2680910 | 2.902 | RNA binding, nucleic acid binding | RNA processing | Lupus La protein; RNA recognition motif, RNP-1 | |
| 2274818 | 2.711 | nucleic acid binding | | RNA recognition motif, RNP-1 | |
| 2675547 | 2.436 | | | | |
| 2695463 | 2.293 | | | | |
| 2753720 | 8.177 | monooxygenase activity, oxidoreductase activity | electron transport, aromatic compound metabolism, metabolism | Monooxygenase, FAD-binding; Aromatic-ring hydroxylase | |
| 2599778 | 6.176 | catalytic activity, oxidoreductase activity | | UBA/THIF-type NAD/FAD binding fold; FAD Dependent reductase | |
| 2550915 | 6.157 | catalytic activity | metabolism | Aminotransferase, class IV | |
| 2668007 | 6.125 | catalytic activity, binding | metabolism | Spermine synthase | |
| 2466833 | 5.997 | catalytic activity, FAD binding, oxidoreductase activity | | FAD-binding, type 2 | |
| 2436028 | 5.561 | chorismate synthase activity | aromatic amino acid family biosynthesis | Chorismate synthase | |
| 2667549 | 5.115 | monooxygenase activity, heme binding, iron ion binding | electron transport | Cytochrome P450 | |
| 2199166 | 4.912 | amino acid transporter activity | amino acid transporter, transport | Amino acid/polyamine transporter I | |
| 2472473 | 4.703 | transferase activity, transferring alkyl or aryl (other than methyl) groups pyridoxal phosphate binding | amino acid metabolism | Cys/Met metabolism, pyridoxal phosphate-dependent enzyme; O-acetylhomoserine/O-acetylserine sulfhydrylase | |
| 2676550 | 4.619 | transferase activity, transferring nitrogenous groups pyridoxal phosphate binding | biosynthesis | Aminotransferase, class I and II | |
| 2603325 | 4.610 | oxidoreductase activity, acting on the CH-CH group of donors, transition metal ion binding, heme binding, acyl-CoA dehydrogenase activity | electron transport, metabolism | Acyl-CoA oxidase/dehydrogenase, type 1 | |
| 2498267 | 4.435 | copper ion binding, amine oxidase activity, quinone binding | amine metabolism | Copper amine oxidase | |
| 2437587 | 4.273 | phosphoribosylanthranilate isomerase activity, catalytic activity, anthranilate synthase activity, indole-3-glycerol-phosphate synthase activity | glutamine metabolism, biosynthesis, tryptophan metabolism, metabolism | N-(5'phosphoribosyl) anthranilate isomerase (PRAI); Glutamine amidotransferase superfamily; Anthranilate synthase component II/delta crystallin; Ribulose-phosphate binding barrel | |
| 2680579 | 4.187 | catalytic activity, binding, oxidoreductase activity | amino acid metabolism, metabolism | Glutamate/phenylalanine/leucine/valine dehydrogenase | |
| 2599439 | 4.166 | monooxygenase activity, NADH dehydrogenase | electron transport | Cytochrome P450 | |

| | | | | | |
|---|---|---|---|---|---|
| | | (ubiquinone) activity, heme binding, iron ion binding, electron carrier activity | | | |
| 2601300 | 3.854 | | | | |
| 2552181 | 3.813 | oxidoreductase activity | electron transport | Taurine catabolism dioxygenase TauD/TfdA | |
| 1188538 | 3.790 | arginase activity, hydrolase activity, acting on carbon-nitrogen (but not peptide) bonds, in linear amidines metal ion binding | arginine catabolism | Arginase; Ureohydrolase | Metabolism |
| 2595827 | 3.602 | catalytic activity, binding, oxidoreductase activity | metabolism | Aldehyde dehydrogenase | |
| 2679197 | 3.255 | hydrolase activity, histone deacetylase activity | histone deacetylation | Metallophosphoesterase; Histone deacetylase | |
| 2600607 | 3.153 | | | | |
| 2680505 | 3.011 | monooxygenase activity, heme binding, iron ion binding | electron transport | Cytochrome P450 | |
| 2600367 | 2.942 | FAD binding, iron ion binding, oxidoreductase activity | electron transport | Ferric reductase-like transmembrane component | |
| 2674336 | 2.799 | glycolipid 2-alpha-mannosyltransferase activity | protein amino acid glycosylation | Glycosyl transferase, family 15 | |
| 2637911 | 2.778 | aldehyde dehydrogenase [NAD(P)] activity, oxidoreductase activity | aldehyde metabolism, metabolism | Aldehyde dehydrogenase | |
| 2681708 | 2.756 | catalytic activity, 3-phosphoshikimate 1-carboxyvinyltransferase activity, hydroxymethylglutaryl-CoA reductase (NADPH) activity, shikimate 5-dehydrogenase activity, shikimate kinase activity, 3-dehydroquinate dehydratase activity, transferase activity, transferring alkyl or aryl (other than methyl) groups, 3-dehydroquinate synthase activity, binding, coenzyme binding, ATP binding | coenzyme A metabolism, aromatic amino acid family biosynthesis, metabolism | RNA 3'-terminal phosphate cyclase/enolpyruvate transferase, alpha/beta; 3-phosphoshikimate 1-carboxyvinyltransferase, subgroup; Hydroxymethylglutaryl-CoA reductase, class I/II, catalytic; Quinate/shikimate 5-dehydrogenase/glutamyl-tRNA reductase; Shikimate-5-dehydrogenase | |
| 2569477 | 2.634 | catalytic activity, binding, coenzyme binding | metabolism | NAD-dependent epimerase/dehydratase | |
| 2510648 | 2.632 | catalytic activity, hydrolase activity, zinc ion binding | | Cytidine deaminase-like | |
| 2550951 | 2.621 | catalytic activity, binding | metabolism | NAD(P)-binding | |
| 2700718 | 2.482 | transaminase activity, pyridoxal phosphate binding | arginine metabolism | Aminotransferase class-III; Acetylornithine and succinylornithine aminotransferase | |
| 2676524 | 2.426 | deaminase activity, AMP deaminase activity | purine ribonucleoside monophosphate, biosynthesis | Adenosine/AMP deaminase | |
| 2690581 | 2.375 | catalytic activity, calcium ion binding, alpha-amylase activity | carbohydrate metabolism | Glycoside hydrolase family 13 | |

| | | | | | |
|---|---|---|---|---|---|
| 2596075 | 2.304 | nucleic acid binding, pyridoxal phosphate binding | cysteine metabolism, metabolism | RNA recognition motif, RNP-1; Aminotransferase, class V/Cysteine desulphurase | |
| 2258133 | 2.290 | metabolism | | Aminotransferase, class V/Cysteine desulphurase | |
| 2705811 | 2.217 | catalytic activity, ATP binding | fructose 2,6-bisphosphate metabolism | Fructose-2,6-bisphosphatase | |
| 2680003 | 2.182 | catalytic activity, ammonia ligase activity, ammonia-lyase activity | biosynthesis, phenylalanine catabolism | L-Aspartase-like; Phenylalanine/histidine ammonia-lyase | |
| 2525947 | 6.940 | nucleic acid binding | | Polynucleotidyl transferase, Ribonuclease H fold | |
| 2600178 | 5.840 | beta-catenin binding | development | Beta-catenin-interacting ICAT | |
| 2678866 | 5.007 | | | | |
| 2675600 | 4.970 | hydrolase activity | | NUDIX hydrolase | |
| 2677822 | 4.210 | ligase activity, ATP binding | mismatch repair | Carboxyl transferase; Acetyl-coenzyme A carboxyltransferase | |
| 2702302 | 3.847 | nucleic acid binding | | Polynucleotidyl transferase, Ribonuclease H fold | |
| 2691250 | 3.397 | nucleic acid binding, RNA binding, zinc ion binding | | Zinc finger, CCCH-type; RNA recognition, region 1 | |
| 2583307 | 2.414 | | | | |
| 2753299 | 6.202 | protein kinase activity, protein-tyrosine kinase activity, protein serine/threonine kinase activity, ATP binding | protein amino acid phosphorylation | Protein kinase; Serine/threonine protein kinase-related; Protein kinase ATP binding | |
| 2750523 | 5.600 | hydrolase activity | metabolism | Lipase, GDXG, active site | |
| 2640974 | 5.087 | | | | |
| 2732602 | 4.615 | peptidase activity | proteolysis and peptidolysis | Peptidase M28 | |
| 2192995 | 4.322 | | | | |
| 2101614 | 4.293 | | | | |
| 2009173 | 3.812 | | | | |
| 2721824 | 3.121 | protein binding | cell wall organization and biogenesis, protein localization | Vacuolar protein sorting-associated protein | Cellular processes and signaling |
| 2558782 | 2.919 | prolyl oligopeptidase activity, serine-type peptidase activity, serine-type endopeptidase activity | proteolysis and peptidolysis | Peptidase S9A, prolyl oligopeptidase | |
| 2502077 | 2.830 | | | | |
| 2603157 | 2.808 | aspartic-type endopeptidase activity, transporter activity, pepsin A activity | oligopeptide transport, proteolysis and peptidolysis | Peptidase aspartic, catalytic | |
| 2570847 | 2.466 | | | | |
| 2675881 | 2.432 | protein binding, GTP binding, nucleic acid binding, GTPase activity | nucleocytoplasmic transport, signal transduction, small GTPase mediated signal transduction, protein transport, intracellular, protein transport | Ras small GTPase, Ras type; Ras small GTPase, Rho type; Ras small GTPase, Rab type; Ran GTPase; Small GTP-binding protein | |

| | | | | | |
|---|---|---|---|---|---|
| 2674662 | 2.278 | microtubule binding | microtubule cytoskeleton organization and biogenesis | HOOK | |
| 2541149 | 10.036 | | | | |
| 2745841 | 8.559 | | | | |
| 2367013 | 7.346 | | | | |
| 2618740 | 6.818 | | | | |
| 2470835 | 6.425 | | | | |
| 2750840 | 6.396 | | | | |
| 2295360 | 6.180 | | | | |
| 2638754 | 5.870 | FAD binding, pyranose oxidase activity, oxidoreductase activity, acting on CH-OH group of donors, oxidoreductase activity | glucose metabolism | Glucose-methanol-choline oxidoreductase; Pyranose oxidase; FAD dependent oxidoreductase | |
| 2503584 | 5.628 | | | | |
| 2549292 | 5.549 | | | | |
| 2750895 | 5.513 | | | | |
| 2597797 | 5.511 | N-acetyltransferase activity | metabolism | GCN5-related N-acetyltransferase | |
| 2748821 | 5.506 | catalytic activity hydrolase activity, acting on carbon-nitrogen (but not peptide) bonds | carbohydrate metabolism | Glycoside hydrolase/deacetylase, beta/alpha-barrel; Polysaccharide deacetylase | |
| 2534073 | 5.483 | | | | |
| 2317702 | 5.408 | nucleic acid binding, zinc ion binding | | Zinc finger, C2H2-type | |
| 2625719 | 5.385 | aminopeptidase activity, prolyl aminopeptidase activity | proteolysis and peptidolysis | Peptidase S33, prolyl aminopeptidase | |
| 2579584 | 5.267 | subtilase activity, serine-type peptidase activity | proteolysis and peptidolysis | | |
| 2476158 | 5.141 | subtilase activity, serine-type peptidase activity | proteolysis and peptidolysis | Peptidase S8 and S53, subtilisin, kexin, sedolisin | |
| 2598518 | 4.447 | | | | |
| 2602331 | 4.438 | | | | |
| 2603514 | 4.416 | nucleic acid binding | | RNA recognition motif, RNP-1 | |
| 2642225 | 4.186 | | | | |
| 2678339 | 4.004 | | | | |
| 2685524 | 3.918 | catalytic activity, binding, zinc ion binding, oxidoreductase activity | metabolism | Alcohol dehydrogenase, zinc-containing; Aldo/keto reductase | |
| 2600246 | 3.796 | metalloendopeptidase activity, hydrolase activity, hydrolyzing O-glycosyl compounds | carbohydrate metabolism, proteolysis and peptidolysis | Peptidase M35, deuterolysin; Glycoside hydrolase, family 31 | |
| 2586726 | 3.788 | | | | |
| 2680606 | 3.733 | | | | |
| 2628500 | 3.671 | | | | |
| 1105701 | 3.670 | | | | |
| 2599691 | 3.592 | | | | |
| 2701101 | 3.565 | | | | |
| 2541924 | 3.461 | | | | |
| 2677655 | 3.428 | catalytic activity, protein kinase activity, protein serine/threonine kinase activity, binding, ATP binding, protein-tyrosine kinase activity | protein amino acid phosphorylation, metabolism | Protein kinase; Serine/threonine protein kinase-related; Protein kinase ATP binding, conserved site | |

| | | | | | |
|---|---|---|---|---|---|
| 2181773 | 3.387 | | | | |
| 2685946 | 3.358 | oxidoreductase activity | electron transport | Amine oxidase | |
| 2602828 | 3.313 | catalytic activity, hydrolase activity, acting on carbon-nitrogen (but not peptide) bonds | carbohydrate metabolism | Polysaccharide deacetylase; Glycoside hydrolase/deacetylase, beta/alpha-barrel | |
| 2179879 | 3.297 | copper ion binding, oxidoreductase activity | | Multicopper oxidase, copper-binding site | |
| 2707024 | 3.282 | structural constituent of ribosome | protein biosynthesis | Ribosomal protein L19/L19e | |
| 2587232 | 3.186 | sugar porter activity, transporter activity | carbohydrate transport, transport | Sugar transporter; General substrate transporter; Major facilitator superfamily | |
| 2581083 | 3.064 | RNA binding, nucleic acid binding | mRNA metabolism | Polyadenylate-binding protein; RNA recognition motif, RNP-1 | |
| 2684270 | 3.037 | | | | |
| 2593925 | 3.017 | 3-deoxy-7-phosphoheptulonate synthase activity | aromatic amino acid family biosynthesis | DAHP synthetase, class II | |
| 2491078 | 3.007 | transferase activity, transferring hexosyl groups | carbohydrate biosynthesis | Glycosyl transferase, family 8 | |
| 2480617 | 2.999 | transcription factor activity, signal transducer activity | signal transduction, regulation of transcription, DNA-dependent | STAT transcription factor, coiled coil | |
| 2602724 | 2.824 | | | | |
| 2442813 | 2.770 | | | | |
| 2527041 | 2.720 | nucleic acid binding, zinc ion binding | | Zinc finger, CCHC-type | |
| 2679354 | 2.511 | catalytic activity, carbamoyl-phosphate synthase activity, ATP binding | metabolism, nitrogen metabolism | Carbamoyl phosphate synthetase, large subunit, ATP-binding; ATP-grasp fold; PreATP-grasp-like fold | |
| 2700082 | 2.492 | zinc ion binding | | Zinc finger, MYND-type | Poorly characterized |
| 2751139 | 2.417 | | | | |
| 2529447 | 2.414 | hydrolase activity, hydrolyzing O-glycosyl compounds | carbohydrate metabolism | Glycoside hydrolase, family 5 | |
| 2687268 | 2.360 | | | | |
| 2517501 | 2.276 | oxidoreductase activity, acting on single donors with incorporation of molecular oxygen, incorporation of two atoms of oxygen, catalytic activity, ferric iron binding, iron ion binding | aromatic compound metabolism | Intradiol ring-cleavage dioxygenase | |
| 2535493 | 2.211 | hydrolase activity, hydrolyzing O-glycosyl compounds | carbohydrate metabolism | Glycoside hydrolase, family 5 | |
| 2661009 | 2.130 | RNA binding, nucleic acid binding | mRNA metabolism | Polyadenylate binding protein, human types 1, 2, 3, 4; RNA recognition motif, RNP-1 | |
| 2449841 | 2.006 | protein binding | | BTB/POZ fold | |

## Table S16. Induced proteins in primordia vs. monokaryon 4-39

| Schco3 ID | Fold change | Go Desc molecular function | Go Desc Biological Process | Interpro Desc | KOG Group |
|---|---|---|---|---|---|
| 2695463 | 6.264 | | | | |
| 2110112 | 5.312 | | | Auxin efflux carrier | |
| 2660823 | 5.052 | metalloendopeptidase activity | proteolysis and peptidolysis | Peptidase M35, deuterolysin | |
| 2717187 | 4.703 | | | | |
| 2599835 | 4.469 | catalytic activity, binding, carbohydrate binding, transcriptional repressor activity | carbohydrate catabolism, regulation of nitrogen utilization, metabolism | Carbohydrate-binding family 9/cellobiose dehydrogenase, cytochrome | |
| 2680910 | 4.046 | RNA binding, nucleic acid binding | RNA processing | Lupus La protein; RNA recognition motif, RNP-1 | |
| 2680216 | 3.712 | nucleic acid binding, zinc ion binding | | Zinc finger, C2H2-type | |
| 2584235 | 3.452 | phosphoric monoester hydrolase activity | metabolism | HAD-superfamily hydrolase, subfamily IB, PSPase-like | |
| 2680499 | 3.430 | cytochrome-c oxidase activity | electron transport | Cytochrome c oxidase, subunit Vib | |
| 2289565 | 3.335 | metalloendopeptidase activity, metallopeptidase activity, zinc ion binding | proteolysis and peptidolysis | Peptidase M48, Ste24p | |
| 2678477 | 3.259 | monooxygenase activity, iron ion binding, sugar porter activity, caspase activity, transporter activity, heme binding, hydrolase activity, oxidoreductase activity | electron transport, carbohydrate transport, proteolysis and peptidolysis, transport | Cytochrome P450; Sugar transporter; Peptidase C14, caspase catalytic; Major facilitator superfamily; Sugar transporter, conserved site | |
| 2601623 | 3.213 | catalytic activity, carbamoyl-phosphate synthase activity, hydrolase activity | glutamine metabolism, biosynthesis, nitrogen metabolism | Anthranilate synthase component II/delta crystallin, Carbamoyl phosphate synthase, GATase region, Glutamine amidotransferase class-I | |
| 2675502 | 2.898 | | | | |
| 2748760 | 2.769 | | | | |
| 2596945 | 2.547 | hydrogen-transporting ATPase activity, rotational mechanism, protein binding, hydrogen-transporting ATP synthase activity, rotational mechanism, zinc ion binding | ATP synthesis coupled proton transport | ATPase, F1 complex, OSCP/delta subunit | |
| 2674509 | 2.475 | actin binding, translation initiation factor activity | cytoskeleton organization and biogenesis, translational initiation, protein biosynthesis | Adenylate cyclase-associated CAP; Translation initiation factor SUI1; Translation initiation factor SUI1 | |
| 2611195 | 2.390 | | | | |
| 2587232 | 2.379 | sugar porter activity, transporter activity | carbohydrate transport, transport | General substrate transporter, Major facilitator superfamily, Sugar transporter, conserved site | |
| 2599506 | 2.192 | C-8 sterol isomerase activity | ergosterol biosynthesis | ERG2 and sigma1 receptor-like | |
| 2602357 | 2.174 | protein binding | amino acid transport, transport | Amino acid permease, conserved site; BTB/POZ fold | |

| | | | | | |
|---|---|---|---|---|---|
| 2274818 | 2.156 | nucleic acid binding | | RNA recognition motif, RNP-1 | |
| 2195169 | 2.152 | | | | |
| 2595331 | 2.135 | protein binding, protein kinase activity, protein serine/threonine kinase activity, binding, ATP binding | protein amino acid phosphorylation, RNA metabolism | Protein kinase; Serine/threonine protein kinase-related; MIF4G-like, type 3 | |
| 2675547 | 2.115 | | | | |
| 2681478 | 2.107 | | nucleic acid binding, zinc ion binding | | |
| 2753720 | 8.250 | monooxygenase activity; oxidoreductase activity | electron transport, aromatic compound metabolism, metabolism | Monooxygenase; Aromatic-ring hydroxylase | |
| 1188538 | 7.803 | arginase activity, hydrolase activity, acting on carbon-nitrogen (but not peptide) bonds, in linear amidines | arginine catabolism | Arginase; Ureohydrolase | Metabolism |
| 2498267 | 6.885 | copper ion binding, amine oxidase activity, quinone binding | amine metabolism | Copper amine oxidase | |
| 2199166 | 6.633 | amino acid transporter activity | amino acid transport, transport | Amino acid/polyamine transporter I | |
| 2675740 | 6.275 | | | | |
| 2599778 | 6.119 | catalytic activity, oxidoreductase activity | | UBA/THIF-type NAD/FAD binding fold; FAD dependent oxidoreductase | |
| 2603325 | 6.011 | oxidoreductase activity, acting on the CH-CH group of donors, transition metal ion binding, acyl-CoA dehydrogenase activity | electron transport, metabolism | Acyl-CoA oxidase/dehydrogenase; Cytochrome b5 | |
| 2668007 | 5.979 | catalytic activity, binding | metabolism | Spermine synthase | |
| 2550915 | 5.952 | catalytic activity | metabolism | Aminotransferase, class IV | |
| 2750840 | 5.937 | | | | |
| 2569477 | 5.917 | catalytic activity, binding, coenzyme binding | metabolism | NAD-dependent epimerase/dehydratase | |
| 2436028 | 5.537 | chorismate synthase activity | aromatic amino acid family biosynthesis | Chorismate synthase | |
| 2680003 | 5.432 | catalytic activity, ammonia ligase activity, ammonia-lyase activity | biosynthesis, phenylalanine catabolism | Phenylalanine/histidine ammonia-lyase; L-Aspartase-like | |
| 2472473 | 5.423 | transferase activity, transferring alkyl or aryl (other than methyl) groups, pyridoxal phosphate binding | amino acid metabolism | O-acetylhomoserine/O-acetylserine sulfhydrylase; Cys/Met metabolism, pyridoxal phosphate-dependent enzyme | |
| 2258133 | 5.348 | metabolism | | Aminotransferase, class V/Cysteine desulphurase | |
| 2676550 | 5.343 | transferase activity, transferring nitrogenous groups, pyridoxal phosphate binding | biosynthesis | Aminotransferase, class I and II | |
| 2680505 | 4.802 | monooxygenase activity, heme binding, iron ion binding | electron transport | Cytochrome P450 | |
| 2597073 | 4.770 | amino acid transport, transport | | Amino acid permease, conserved site | |

| | | | | | |
|---|---|---|---|---|---|
| 2466833 | 4.512 | catalytic activity, FAD binding, oxidoreductase activity | | FAD-binding, type 2 | |
| 2595653 | 4.408 | kynureninase activity, pyridoxal phosphate binding, oxidoreductase activity | tryptophan catabolism, nicotinamide adenine dinucleotide biosynthesis, metabolism | FAD dependent oxidoreductase; Kynureninase | |
| 2510648 | 4.404 | catalytic activity, hydrolase activity, zinc ion binding | | Cytidine deaminase-like | |
| 2700718 | 4.277 | transaminase activity, pyridoxal phosphate binding | arginine metabolism | Aminotransferase class-III; Acetylornithine and succinylornithine aminotransferase | |
| 2705811 | 4.024 | catalytic activity, ATP binding | fructose 2,6-bisphosphate metabolism | Fructose-2,6-bisphosphatase | |
| 2700638 | 3.900 | purine-nucleoside phosphorylase activity, transferase activity, transferring pentosyl groups | nucleobase, nucleoside, nucleotide and nucleic acid metabolism | Inosine guanosine and xanthosine phosphorylase; Purine phosphorylase, family 2 | |
| 2674336 | 3.725 | glycolipid 2-alpha-mannosyltransferase activity | protein amino acid glycosylation | Glycosyl transferase, family 15 | |
| 2681970 | 3.449 | catalytic activity | metabolism | AMP-dependent synthetase and ligase | |
| 2596642 | 3.424 | nucleoside triphosphatase activity, nucleotide binding, ATPase activity, coupled to transmembrane movement of substances, ATPase activity, ATP binding | transport | AAA+ ATPase, core; ABC transporter, transmembrane region | |
| 2552181 | 3.419 | oxidoreductase activity | electron transport | Taurine catabolism dioxygenase TauD/TfdA | |
| 2676524 | 3.404 | deaminase activity, AMP deaminase activity | purine ribonucleoside monophosphate biosynthesis | AMP deaminase | |
| 2643580 | 3.329 | transporter activity, citrate transporter activity | sodium ion transport, citrate transport | Sodium/sulphate symporter; Divalent ion symporter | |
| 2476486 | 3.313 | 3-isopropylmalate dehydratase activity | metabolism, leucine biosynthesis | Aconitase/3-isopropylmalate dehydratase large subunit, alpha/beta/alpha | |
| 2679876 | 3.201 | catalytic activity, cofactor binding, transporter activity, binding | transport, metabolism | AMP-dependent synthetase and ligase; Phosphopantetheine-binding; Major facilitator superfamily | |
| 2536606 | 3.191 | DNA binding, pantoate-beta-alanine ligase activity, DNA-directed RNA polymerase activity | transcription, pantothenate biosynthesis | DNA-directed RNA polymerase, bacteriophage type; Pantoate-beta-alanine ligase | |
| 2595640 | 3.139 | asparagine synthase (glutamine-hydrolyzing) activity | asparagine biosynthesis, metabolism | Asparagine synthase | |
| 2443639 | 3.099 | lactoylglutathione lyase activity | carbohydrate metabolism | Glyoxalase I | |
| 2678898 | 3.024 | guanyl nucleotide binding, carbon-nitrogen ligase activity, with glutamine as amido-N-donor, signal transducer activity | signal transduction, G-protein coupled receptor protein signaling pathway | Guanine nucleotide binding protein (G-protein), alpha subunit; Amidase signature enzyme; G protein alpha subunit, helical insertion | |

| | | | | | |
|---|---|---|---|---|---|
| 2597924 | 3.006 | catalytic activity, binding, oxidoreductase activity | metabolism | Short-chain dehydrogenase /reductase SDR; Glucose/ribitol dehydrogenase; Flavoprotein | |
| 1118808 | 2.936 | catalytic activity | | Phosphatidic acid phosphatase type 2/haloperoxidase | |
| 2597675 | 2.829 | catalytic activity, transcription factor activity, dihydroxy-acid dehydratase activity, zinc ion binding | branched chain family amino acid biosynthesis, regulation of transcription, DNA-dependent, metabolism | Dihydroxy-acid and 6-phosphogluconate dehydratase; Fungal transcriptional regulatory protein; Dihydroxy-acid dehydratase | |
| 2440737 | 2.770 | hydrolase activity | | Amidohydrolase 1 | |
| 2578054 | 2.703 | cytochrome-c oxidase activity | electron transport | Cytochrome c oxidase, subunit Vb | |
| 2091788 | 2.659 | NADH dehydrogenase activity, NADH dehydrogenase (ubiquinone) activity | electron transport | NADH-ubiquinone oxidoreductase B18 subunit | |
| 2674539 | 2.593 | hydroxymethylglutaryl-CoA reductase (NADPH) activity, oxo-acid-lyase activity, anthranilate synthase activity, coenzyme binding | coenzyme A metabolism, biosynthesis, tryptophan biosynthesis | Hydroxymethylglutaryl-CoA reductase, class I/II, catalytic; Anthranilate synthase component I | |
| 2600532 | 2.592 | DNA binding, transporter activity, hydrolase activity, acting on carbon-nitrogen (but not peptide) bonds, RNA polymerase II transcription factor activity | transport, regulation of transcription | Histone-fold; Major facilitator superfamily; Metal-dependent hydrolase, composite | |
| 2706358 | 2.520 | NAD ADP-ribosyltransferase activity, mevalonate kinase activity, phosphotransferase activity, alcohol group as acceptor, ATP binding, pyridoxal phosphate, binding, kinase activity | amino acid metabolism, isoprenoid biosynthesis, phosphorylation, metabolism | GHMP kinase; Mevalonate kinase | |
| 2494667 | 2.517 | | | ERG4/ ERG24 ergosterol biosynthesis protein | |
| 2531122 | 2.498 | metallopeptidase activity, binding, zinc ion binding | proteolysis and peptidolysis, leukotriene biosynthesis | Peptidase M1, membrane alanine aminopeptidase; Armadillo-type fold | |
| 2525861 | 2.492 | imidazoleglycerol-phosphate dehydratase activity | histidine biosynthesis | Imidazole glycerol-phosphate dehydratase | |
| 2290196 | 2.459 | | | | |
| 2679472 | 2.446 | protein binding, GTP binding, GTPase activity | nucleocytoplasmic transport, signal transduction, small GTPase, mediated signal transduction, protein transport, intracellular protein transport | Ras small GTPase, Ras type; Ras small GTPase, Rho type; Ras small GTPase, Rab type; Ran GTPase; Small GTP-binding protein | |
| 2603078 | 2.395 | ATP binding, nucleotide binding, nucleoside triphosphatase activity, | anti-apoptosis, transport | AAA+ ATPase; ABC transporter, transmembrane region | |

| | | | | | |
|---|---|---|---|---|---|
| | | ATPase activity, ATPase activity, coupled to transmembrane movement of substances | | | |
| 2595827 | 2.277 | catalytic activity, binding, oxidoreductase activity | metabolism | Aldehyde dehydrogenase | |
| 2550951 | 2.240 | | | | |
| 2621172 | 2.199 | | | | |
| 2752605 | 2.120 | N-acetyltransferase activity | metabolism | GCN5-related N-acetyltransferase | |
| 2667262 | 2.085 | hydrolase activity, acting on carbon-nitrogen (but not peptide) bonds, in cyclic amides, dihydroorotase activity | pyrimidine base biosynthesis | Dihydroorotase, conserved site | |
| 2596075 | 2.083 | nucleic acid binding, pyridoxal phosphate binding | cysteine metabolism, metabolism | RNA recognition motif, RNP-1; Cysteine desulphurase | |
| 2600367 | 2.032 | FAD binding, iron ion binding, oxidoreductase activity | electron transport | Ferric reductase-like transmembrane component | |
| 2525947 | 6.847 | nucleic acid binding | | Polynucleotidyl transferase, Ribonuclease H fold | |
| 2600178 | 4.922 | beta-catenin binding | development | Beta-catenin-interacting ICAT | |
| 2678866 | 4.179 | | | | |
| 2699460 | 3.655 | helicase activity, RNA binding, nucleic acid binding, ribonuclease III activity, ATP dependent helicase activity, ATP binding | RNA processing | DNA/ RNA helicase; Ribonuclease III | |
| 2662001 | 3.300 | DNA-directed RNA polymerase activity | transcription | RNA polymerases, N/8 Kd subunits | |
| 2702302 | 3.267 | nucleic acid binding | | Polynucleotidyl transferase, Ribonuclease H fold | |
| 2694333 | 2.991 | nucleic acid binding | | RNA recognition motif, RNP-1 | |
| 2747036 | 2.933 | zinc ion binding | | Zinc finger, MYND-type | |
| 2691250 | 2.850 | nucleic acid binding, RNA binding, zinc ion binding | | Zinc finger, CCCH-type; RNA recognition, region 1 | |
| 2020262 | 2.800 | transferase activity, binding, acyltransferase activity | | Trimeric LpxA-like; Armadillo-type fold | |
| 2528965 | 2.769 | double-stranded RNA binding, RNA binding, ribonuclease III activity | RNA processing | Double-stranded RNA binding; Ribonuclease III | |
| 2628500 | 2.697 | | | | |
| 2598981 | 2.509 | zinc ion binding | | Zinc finger, MYND-type; Batten's disease protein Cln3 | |
| 2676995 | 2.435 | nucleic acid binding, zinc ion binding | | Zinc finger, CCCH-type | |
| 2680463 | 2.346 | helicase activity, catalytic activity, CTP synthase activity, nucleic acid binding, ATP dependent helicase activity, ATP binding | pyrimidine nucleotide biosynthesis | DNA/RNA helicase; Glutamine amidotransferase; CTP synthase | |
| 2543008 | 2.300 | methionine-tRNA ligase activity, nucleotide binding, tRNA ligase activity, ATP binding | methionyl-tRNA aminoacylation, amino acid activation, protein biosynthesis | Methionyl-tRNA synthetase, class Ia; Aminoacyl-tRNA synthetase, class I, conserved site | |
| 2602443 | 2.205 | nucleic acid binding | | RNA recognition motif, RNP-1 | |

| | | | | | |
|---|---|---|---|---|---|
| 1137889 | 2.132 | RNA processing, mRNA processing | | RNA-processing protein, HAT helix | |
| 2009173 | 9.152 | | | | |
| 2192995 | 7.629 | | | | |
| 2702602 | 6.609 | calcium ion binding | | Calcium-binding EF-hand | |
| 2732602 | 5.615 | DNA binding, DNA-directed RNA polymerase activity | transcription | DNA-directed RNA polymerase, M/15 kDa subunit | |
| 2750523 | 5.454 | hydrolase activity | metabolism | Lipase, GDXG, active site | |
| 2497887 | 5.167 | peptidase activity | proteolysis and peptidolysis | Peptidase M28 | |
| 2101614 | 4.704 | | | | |
| 2558782 | 4.206 | prolyl oligopeptidase activity, serine-type peptidase activity, serine-type endopeptidase activity | proteolysis and peptidolysis | Peptidase S9A, prolyl oligopeptidase | |
| 2753299 | 4.124 | protein kinase activity, protein-tyrosine kinase activity, protein serine/threonine kinase activity, ATP binding | protein amino acid phosphorylation | Tyrosine protein kinase; Serine/threonine protein kinase; Protein kinase ATP binding, conserved site | |
| 2674662 | 4.066 | microtubule binding | microtubule cytoskeleton organization and biogenesis | HOOK | |
| 2596898 | 3.581 | oxidoreductase activity | metabolism | Aldehyde dehydrogenase, conserved site | |
| 2572877 | 3.554 | peptidase activity | proteolysis and peptidolysis | Peptidase M28 | |
| 2570847 | 3.503 | | | | |
| 2596052 | 3.377 | peptidase activity | proteolysis and peptidolysis | Peptidase M28 | |
| 2747374 | 2.601 | catalytic activity, acid phosphatase activity, ATP binding | | ATP-grasp fold; Histidine acid phosphatase | |
| 2674486 | 2.334 | catalytic activity, transferase activity, transferring nitrogenous groups, transaminase activity, pyridoxal phosphate binding | biosynthesis, amino acid metabolism | Aminotransferase | |
| 2680842 | 2.287 | steroid metabolism | | Oxysterol-binding protein | |
| 2678033 | 2.212 | | | | |
| 2558600 | 2.181 | protein kinase activity, protein serine/threonine kinase activity, ATP binding | protein amino acid phosphorylation | Protein kinase; Serine/threonine protein kinase-related | |
| 2677252 | 2.148 | oligosaccharyl transferase activity | protein amino acid glycosylation | Oligosaccharyl transferase, STT3 subunit | |
| 2600797 | 2.098 | ER retention sequence binding | protein-ER retention | ER lumen protein retaining receptor | |
| 2678792 | 2.083 | nucleoside triphosphatase activity, nucleotide binding, ATP binding | | AAA ATPase, core | |
| 2545891 | 2.079 | calcium ion binding | | Calcium-binding EF-hand | |
| 2749150 | 9.671 | | | Allergen V5/Tpx-1 related | |
| 2541149 | 9.036 | | | | |
| 2745841 | 7.858 | | | | |
| 2603198 | 7.250 | | | | |
| 2367013 | 7.049 | | | | |
| 2549292 | 6.719 | | | | |

| | | | | | |
|---|---|---|---|---|---|
| 2470835 | 6.569 | | | | |
| 2579584 | 6.404 | subtilase activity, serine-type peptidase activity | proteolysis and peptidolysis | Peptidase S8 and S53, subtilisin, kexin, sedolisin | |
| 2295360 | 6.300 | | | | |
| 2308933 | 6.241 | | | Allergen V5/Tpx-1 related | |
| 2638754 | 6.009 | FAD binding oxidoreductase activity, acting on CH-OH group of donors, oxidoreductase activity | glucose metabolism | Glucose-methanol-choline oxidoreductase; Pyranose oxidase; Glucose-methanol-choline oxidoreductase; FAD dependent oxidoreductase | |
| 2491078 | 5.861 | transferase activity, transferring hexosyl groups | carbohydrate biosynthesis | Glycosyl transferase, family 8 | |
| 2618740 | 5.456 | | | | |
| 2534073 | 5.411 | | | | |
| 2677004 | 5.314 | | | | |
| 2317702 | 5.238 | nucleic acid binding, zinc ion binding | | Zinc finger, C2H2-type | |
| 2598518 | 5.139 | | | | |
| 2667549 | 5.018 | monooxygenase activity, heme binding, iron ion binding | electron transport | Cytochrome P450 | |
| 2597797 | 5.005 | N-acetyltransferase activity | | GCN5-related N-acetyltransferase | |
| 2506333 | 4.857 | | | | |
| 2678339 | 4.812 | | | | |
| 2700082 | 4.768 | zinc ion binding | | Zinc finger, MYND-type | Poorly characterized |
| 2602828 | 4.382 | catalytic activity, hydrolase activity, acting on carbon-nitrogen (but not peptide) bonds | carbohydrate metabolism | Polysaccharide deacetylase; Glycoside hydrolase/deacetylase, beta/alpha-barrel | |
| 2685946 | 4.369 | oxidoreductase activity | electron transport | Amine oxidase | |
| 2449841 | 4.224 | protein binding | | BTB/POZ fold | |
| 2602716 | 4.162 | peroxidase activity, heme binding | electron transport, response to oxidative stress | Haem peroxidase | |
| 2680606 | 4.104 | | | | |
| 2503584 | 4.096 | | | | |
| 2676242 | 4.089 | DNA binding, NAD-dependent histone deacetylase activity, zinc ion binding | chromatin silencing, regulation of transcription, DNA-dependent, protein amino acid deacetylation | NAD-dependent histone deacetylase, silent information regulator Sir2 | |
| 2586726 | 3.748 | | | | |
| 2442813 | 3.519 | | | | |
| 2541924 | 3.407 | | | | |
| 2599691 | 3.329 | | | | |
| 2639888 | 3.286 | | | | |
| 2525766 | 3.281 | | | | |
| 2750895 | 3.270 | | | | |
| 2625719 | 3.236 | aminopeptidase activity, prolyl aminopeptidase activity | proteolysis and peptidolysis | Peptidase S33, tricorn interacting factor 1 | |
| 2179879 | 3.146 | copper ion binding, oxidoreductase activity | | Multicopper oxidase, type 2 | |

| | | | | | |
|---|---|---|---|---|---|
| 2593925 | 3.085 | 3-deoxy-7-phosphoheptulonate synthase activity | aromatic amino acid family biosynthesis | DAHP synthetase, class II | |
| 2018047 | 2.996 | peroxidase activity | electron transport | Chloroperoxidase | |
| 2667748 | 2.985 | | | | |
| 2753486 | 2.970 | | | | |
| 2597288 | 2.851 | FAD binding, oxidoreductase activity, acting on CH-OH group of donors | | Glucose-methanol-choline oxidoreductase | |
| 1199149 | 2.820 | | | | |
| 2677660 | 2.804 | zinc ion binding | | Zinc finger, B-box | |
| 2684270 | 2.660 | | | | |
| 2675539 | 2.659 | transcription factor activity, nucleic acid binding, zinc ion binding | regulation of transcription, DNA-dependent | Fungal transcriptional regulatory protein; Zinc finger, CCCH-type | |
| 2704482 | 2.656 | | | | |
| 2595539 | 2.640 | | | | |
| 2748821 | 2.600 | catalytic activity, hydrolase activity, acting on carbon-nitrogen (but not peptide) bonds | carbohydrate metabolism | Polysaccharide deacetylase; Glycoside hydrolase/deacetylase, beta/alpha-barrel | |
| 2679197 | 2.519 | hydrolase activity, histone deacetylase activity | histone deacetylation | Metallophosphoesterase; Histone deacetylase | |
| 2601373 | 2.474 | | | | |
| 2516775 | 2.474 | | | | |
| 2545442 | 2.445 | hydrolase activity, hydrolyzing O-glycosyl compounds | carbohydrate metabolism | Glycoside hydrolase, family 3 | |
| 2707024 | 2.443 | structural constituent of ribosome | protein biosynthesis | Ribosomal protein L19/L19e | |
| 2746514 | 2.421 | ATP binding, zinc ion binding, DNA binding, NAD synthase (glutamine-hydrolyzing) activity, hydrolase activity, acting on carbon-nitrogen (but not peptide) bonds | transcription, nicotinamide adenine dinucleotide biosynthesis, nitrogen metabolism | Fungal specific transcription factor; Nitrilase/cyanide hydratase and apolipoprotein N-acyltransferase; Fungal specific transcription factor | |
| 2527041 | 2.393 | nucleic acid binding, zinc ion binding | | Zinc finger, CCHC-type | |
| 2517501 | 2.383 | oxidoreductase activity, acting on single donors with incorporation of molecular oxygen, incorporation of two atoms of oxygen, catalytic activity, ferric iron binding, iron ion binding | aromatic compound metabolism | Intradiol ring-cleavage dioxygenase | |
| 2680023 | 2.369 | hydrolase activity, acting on carbon-nitrogen (but not peptide) bonds, chorismate synthase activity, hydrolase activity | aromatic amino acid family biosynthesis | Metal-dependent hydrolase, composite; Chorismate synthase; Amidohydrolase 1 | |
| 2601132 | 2.305 | hydrolase activity, hydrolyzing O-glycosyl compounds, beta-lactamase activity, zinc ion binding | antibiotic catabolism, carbohydrate metabolism | Glycoside hydrolase, family 43; Beta-lactamase, class B, conserved site | |
| 2602724 | 2.280 | | | | |
| 2681580 | 2.269 | nucleoside triphosphatase activity, nucleotide | | AAA+ ATPase, core; ABC transporter, transmembrane region | |

| | | | | | |
|---|---|---|---|---|---|
| | | binding, ATPase activity, ATP binding | | | |
| 2233882 | 2.152 | | | | |
| 2674711 | 2.125 | | | Beta-lactamase-like | |
| 2705231 | 2.094 | hydrolase activity | | NUDIX hydrolase, core | |
| 2681337 | 2.082 | actin binding | cytoskeleton organization and biogenesis | Profilin/allergen | |
| 2639294 | 2.070 | protein modification | | Ubiquitin | |
| 2603514 | 2.050 | nucliec acid binding | | RNA recognition motif, RNP-1 | |
| 2679354 | 2.047 | catalytic activity, carbamoyl-phosphate synthase activity, ATP binding | metabolism, nitrogen metabolism | Carbamoyl phosphate synthetase, large subunit, ATP-binding; ATP-grasp fold; PreATP-grasp-like fold | |

### Table S17. Repressed proteins in primordia vs. monokaryon 12-43

| Schco3 ID | Fold Change | Go Desc molecular function | Go Desc Biological Process | Interpro Desc | KOG Group |
|---|---|---|---|---|---|
| 2492200 | -2.016 | serine-type peptidase activity | proteolysis and peptidolysis | Peptidase S28 | |
| 2110939 | -2.050 | | protein folding | Heat shock protein DnaJ | |
| 2445177 | -2.088 | glutathione transferase activity | metabolism | Glutathione S-transferase, omega-class | |
| 2600099 | -2.104 | oxidoreductase activity | | Redoxin | |
| 2565750 | -2.174 | | | | |
| 2599999 | -2.195 | catalytic activity, binding, protein transporter activity, importin-alpha export receptor activity | cell proliferation, protein-nucleus import, docking, apoptosis, intracellular protein transport | Phosphatidic acid phosphatase type 2/haloperoxidase; Armadillo-type fold; Importin-beta; CAS/CSE | |
| 2572877 | -2.198 | peptidase activity | proteolysis and peptidolysis | Peptidase M28 | |
| 2697890 | -2.202 | protein kinase activity, protein-tyrosine kinase activity, protein serine/threonine kinase activity, ATP binding | intracellular signaling cascade, signal transduction, protein amino acid phosphorylation | Protein kinase; Serine/threonine protein kinase-related; Protein kinase ATP binding, conserved site | |
| 2627084 | -2.371 | hydrolase activity | | Serine/threonine-specific protein phosphatase and bis(5-nucleosyl)-tetraphosphatase; Metallophosphoesterase | |
| 2504769 | -2.393 | catalytic activity | | Prenyltransferase/squalene oxidase | |
| 2599215 | -2.469 | GTPase activity, structural molecule activity, GTP binding | microtubule-based movement, microtubule-based process | Tubulin/FtsZ | |
| 2680703 | -2.470 | glucosylceramidase activity, protein binding, binding | lysosome organization and biogenesis, protein complex assembly, sphingolipid metabolism, intracellular protein transport, | Glycoside hydrolase, family 30; Clathrin/coatomer adaptor, adaptin-like; Armadillo-type fold | |

| | | | | | |
|---|---|---|---|---|---|
| | | | vesicle-mediated transport | | |
| 2640508 | -2.476 | phosphatidylinositol phosphate kinase activity | phosphatidylinositol metabolism | Phosphatidylinositol-4-phosphate 5-kinase | |
| 2545132 | -2.491 | GTPase activity, structural molecule activity, GTP binding | microtubule-based movement, microtubule-based process | Tubulin/FtsZ, C-terminal | |
| 1141646 | -2.532 | GTPase activity, structural molecule activity, GTP binding | microtubule-based movement, microtubule-based process | Tubulin/FtsZ | |
| 2354629 | -2.546 | GTP binding | protein transport, small GTPase mediated signal transduction, intracellular protein transport | ARF/SAR superfamily; Ras GTPase; ADP-ribosylation factor | Cellular processes and signaling |
| 2602808 | -2.592 | protein prenyltransferase activity | protein amino acid prenylation | Protein prenyltransferase, alpha subunit | |
| 2677294 | -2.615 | catalytic activity, binding, coenzyme binding | metabolism | NAD-dependent epimerase/dehydratase | |
| 2674968 | -2.807 | nucleic acid binding, ATP binding, zinc ion binding | | Zinc finger, C2H2-type | |
| 2675738 | -2.850 | intracellular protein transport | | NSF attachment protein | |
| 2249278 | -2.866 | | | | |
| 2127007 | -3.283 | metal ion binding | | Class II aldolase/adducin | |
| 2208671 | -3.370 | actin binding, calcium ion binding | | Actinin-type, actin-binding, conserved site; Calcium-binding EF-hand | |
| 2702602 | -3.431 | calcium ion binding | | Calcium-binding EF-hand | |
| 2678401 | -3.899 | GTPase activity, structural molecule activity, GTP binding | microtubule-based movement, microtubule-based process | Tubulin/FtsZ | |
| 2596185 | -3.949 | protein binding, GTP binding, GTPase activity | nucleocytoplasmic transport, signal transduction, small GTPase mediated signal transduction, protein transport, intracellular protein transport | Ras small GTPase, Ras type; Ras small GTPase, Rho type; Ras small GTPase, Rab type; Ran GTPase; Small GTP-binding protein | |
| 2680097 | -4.134 | acid phosphatase activity, zinc ion binding, transcription factor activity, ARF guanyl-nucleotide exchange factor activity | regulation of transcription, DNA-dependent | Histidine acid phosphatase; Zinc finger, GATA-type; SEC7-like | |
| 2750928 | -2.116 | | | Ribosomal protein S8E | |
| 2467294 | -2.140 | structural constituent of ribosome | protein biosynthesis | Ribosomal protein S15 | |
| 2674535 | -2.153 | protein binding, ATP binding | protein folding | Chaperonin Cpn60/TCP-1 | |
| 2680463 | -2.173 | helicase activity, catalytic activity, CTP synthase activity, nucleic acid | pyrimidine nucleotide biosynthesis | DNA/RNA helicase; Glutamine amidotransferase class-I; CTP synthase | |

| | | | | | |
|---|---|---|---|---|---|
| | | binding, ATP dependent helicase activity, ATP binding | | | |
| 2678650 | -2.335 | structural constituent of ribosome | protein biosynthesis | Ribosomal protein L31e | |
| 2743884 | -2.632 | protein kinase activity, transition metal ion binding, protein-tyrosine kinase activity, protein serine/threonine kinase activity, ATP binding; oxidoreductase activity | protein amino acid phosphorylation | Serine/threonine protein kinase-related; Protein kinase ATP binding, conserved site; Ferritin/ribonucleotide reductase-like | |
| 2681298 | -2.818 | thiamin diphosphokinase activity, ATP binding, nucleotide binding, glycine-tRNA ligase activity, tRNA ligase activity | thiamin diphosphate biosynthesis, amino acid activation, thiamin metabolism, protein biosynthesis, glycyl-tRNA aminoacylation | Glycyl-tRNA synthetase, alpha2 dimer; Aminoacyl-tRNA synthetase, class II (G, H, P and S); Anticodon-binding; Thiamin pyrophosphokinase, catalytic region | |
| 2488899 | -3.097 | structural constituent of ribosome | protein biosynthesis | Ribosomal protein S27e | |
| 2491982 | -3.888 | structural constituent of ribosome, RNA binding | protein biosynthesis | Ribosomal protein L23, conserved site | |
| 2680491 | -4.767 | protein binding, ATPase activity, ATP binding | DNA metabolism | SMCs flexible hinge; ABC transporter-like | |
| 2725570 | -4.790 | | | | |
| 2663340 | -6.082 | structural constituent of ribosome | protein biosynthesis | Ribosomal protein S16 | |
| 2601895 | -7.624 | calcium ion binding, ATP binding | mismatch repair | Calcium-binding EF-hand; DNA mismatch repair protein; ATP-binding region, ATPase-like | |
| 2480368 | -2.059 | metal ion binding, superoxide dismutase activity | superoxide metabolism | Manganese and iron superoxide dismutase | |
| 2680146 | -2.071 | nucleic acid binding, ATP binding, nucleotide binding, nucleoside triphosphatase activity, ATPase activity, ATPase activity, coupled to transmembrane movement of substances | transport | RNA recognition motif, RNP-1; ABC transporter-like; AAA+ ATPase, core | |
| 2675304 | -2.108 | beta-galactosidase activity | carbohydrate metabolism | Glycoside hydrolase, family 35 | |
| 2669280 | -2.167 | transporter activity, binding | transport | Adenine nucleotide translocator 1; Mitochondrial substrate carrier | |
| 2511932 | -2.234 | catalytic activity, binding, oxidoreductase activity | electron transport, metabolism | Oxidoreductase | |
| 2680115 | -2.239 | catalytic activity | | UBA/THIF-type NAD/FAD binding fold | |
| 2504219 | -2.269 | guanyl nucleotide binding, GTP binding, signal transducer activity | signal transduction, G-protein coupled receptor protein signaling pathway | Guanine nucleotide binding protein (G-protein), alpha subunit; Fungal G-protein, alpha subunit | |
| 2601008 | -2.272 | catalytic activity, malate synthase activity, methyltransferase activity | glyoxylate cycle, metabolism | Malate synthase | |
| 2131899 | -2.283 | ATP binding | anion transport | Anion-transporting ATPase | |

| | | | | | |
|---|---|---|---|---|---|
| 2273629 | -2.314 | Protein binding | cell communication | Phox-like | |
| 2620107 | -2.414 | catalytic activity, oxidoreductase activity, acting on CH-OH group of donors, binding, oxidoreductase activity, acting on the CH-OH group of donors, NAD or NADP as acceptor, glycerol-3-phosphate, dehydrogenase (NAD) activity, oxidoreductase activity | glycerol-3-phosphate catabolism, glycerol-3-phosphate metabolism, carbohydrate metabolism, metabolism | NAD-dependent glycerol-3-phosphate dehydrogenase | |
| 2516399 | -2.509 | calcium ion binding, mannosyl-oligosaccharide 1,2-alpha-mannosidase activity | | Glycoside hydrolase, family 47 | |
| 2750135 | -2.512 | glucosamine-6-phosphate deaminase activity | N-acetylglucosamine metabolism, carbohydrate metabolism | Glucosamine-6-phosphate isomerase | |
| 2597083 | -2.730 | acyltransferase activity | metabolism | Phospholipid/glycerol acyltransferase | |
| 2624619 | -2.792 | D-amino-acid oxidase activity, oxidoreductase activity | | D-amino acid oxidase, conserved site; FAD dependent oxidoreductase | |
| 2295738 | -2.795 | metal ion transporter activity, zinc ion transporter activity | zinc ion transport, metal ion transport | Zinc/iron permease | |
| 2551610 | -2.860 | peroxidase activity, ATP binding, nucleoside triphosphatase activity, nucleotide binding, heme binding, ATPase activity, ATPase activity, coupled to transmembrane movement of substances | electron transport, transport, response to oxidative stress | Haem peroxidase; ABC transporter, transmembrane region, type 1; AAA+ ATPase, core | |
| 2535945 | -2.885 | phosphoribosylaminoimidazolesuccinocarboxamide synthase activity | purine nucleotide biosynthesis | SAICAR synthetase | |
| 2675904 | -2.913 | hydrolase activity, hydrolyzing O-glycosyl compounds, oxidoreductase activity | carbohydrate metabolism | Glycoside hydrolase family 2, immunoglobulin-like beta-sandwich; Aldo-ketoreductase | |
| 2599939 | -3.113 | catalytic activity, magnesium ion binding | | Thiamine pyrophosphate enzyme, central region | |
| 2596034 | -3.145 | regulation of signal transduction, response to biotic stimulus | | TAP42-like protein | |
| 2274225 | -3.284 | metal ion binding, superoxide dismutase activity | superoxide metabolism | Manganese and iron superoxide dismutase | |
| 2530104 | -3.304 | oxidoreductase activity | | NADH flavin oxidoreductase/NADH oxidase | |
| 2530747 | -3.514 | | | | |
| 2255769 | -3.548 | | | Translationally controlled tumour-associated TCTP | |
| 2674964 | -3.620 | catalytic activity, protein kinase activity, protein serine/threonine kinase activity, ATP binding | protein amino acid phosphorylation, metabolism | Fumarylacetoacetase; Protein kinase; Serine/threonine protein kinase-related | |
| 2676329 | -3.644 | catalytic, phospho-N-acetylmuramoyl-pentapeptide-transferase | carboxylic acid metabolism, metabolism | Isocitrate lyase and phosphorylmutase; Pyruvate/Phosphoenolpyruvate | |

| | | | | | |
|---|---|---|---|---|---|
| | | activity, isocitrate lyase activity | | kinase, catalytic core; Glycosyl transferase, family 4 | |
| 2312113 | -4.068 | catalytic activity, binding, coenzyme binding | metabolism | NAD-dependent epimerase/dehydratase | |
| 2695732 | -4.118 | transferase activity, transferring nitrogenous groups, pyridoxal phosphate binding | biosynthesis | Aminotransferase, class I and II | |
| 2676596 | -4.313 | catalytic activity, carboxyl- and carbamoyltransferase activity, monooxygenase activity, binding, amino acid binding, oxidoreductase activity | electron transport, amino acid metabolism, aromatic compound metabolism, metabolism | Aspartate/ornithine carbamoyltransferase; Monooxygense; Glucose/ribitol dehydrogenase; Pyridine nucleotide-disulphide oxidoreductase, class I; Aromatic-ring hydroxylase | |
| 2595248 | -4.473 | serine carboxypeptidase activity | proteolysis and peptidolysis | Peptidase S10, serine carboxypeptidase | |
| 2680902 | -4.941 | carboxy-lyase, pyridoxal phosphate binding | carboxylic acid metabolism | Pyridoxal phosphate-dependent decarboxylase | |
| 2552020 | -5.449 | catalytic activity | acetyl-CoA metabolism | Acetyl-CoA hydrolase/transferase | |
| 1152047 | -7.687 | catalytic activity | | Epoxide hydrolase-like | |
| 2714417 | -7.778 | | | | |
| 2469166 | -3.938 | | | | Metabolism |
| 2746421 | -2.012 | GTP binding | protein transport, small GTPase mediated signal transduction | Ras small GTPase, Ras type; Ras small GTPase, Rho type; Ras small GTPase, Rab type; Small GTP-binding protein | |
| 2596927 | -2.081 | nucleic acid binding ATP binding, helicase activity, sugar porter activity, transporter activity, ATP dependent helicase activity | carbohydrate transport, transport | DNA/RNA helicase; General substrate transporter; Major facilitator superfamily; Sugar transporter, conserved site | |
| 2480821 | -2.085 | nucleic acid binding | | RNA recognition motif, RNP-1 | |
| 2749685 | -2.093 | sugar porter activity, transporter activity | carbohydrate transport, transport | General substrate transporter; Major facilitator superfamily; Sugar transporter | |
| 2502834 | -2.129 | hydrolase activity | | Dienelactone hydrolase | |
| 2680350 | -2.170 | transporter activity | transport | Major facilitator superfamily | |
| 2676133 | -2.415 | FAD binding, oxidoreductase activity, acting on CH-OH group of donors | | Glucose-methanol-choline oxidoreductase | |
| 2643023 | -2.446 | FAD binding, oxidoreductase activity, acting on CH-OH group of donors | alcohol metabolism | Glucose-methanol-choline oxidoreductase | |
| 2660812 | -3.126 | oxidoreductase activity | negative regulation of transcription | Flavodoxin/nitric oxide synthase | |
| 2693747 | -3.245 | transporter activity, binding | signal transduction, transport | Lipocalin; Rho GTPase activation protein | |
| 2688236 | -4.049 | catalytic activity, binding, ATP binding, nucleotide binding, tRNA ligase activity, oxidoreductase activity | amino acid activation, protein biosynthesis, metabolism | Aminoacyl-tRNA synthetase, class I; Short-chain dehydrogenase/reductase SDR; Glucose/ribitol dehydrogenase | |
| 2696064 | -4.168 | FAD binding, oxidoreductase activity, acting on CH-OH group of donors | alcohol metabolism | Glucose-methanol-choline oxidoreductase | |

| | | | | | |
|---|---|---|---|---|---|
| 2600127 | -4.626 | | | | |
| 2231044 | -4.682 | | | | |
| 2696773 | -5.174 | | | | |
| 2673553 | -2.005 | catalytic activity, hydrolase activity, acting on carbon-nitrogen (but not peptide) bonds | carbohydrate metabolism | Glycoside hydrolase/deacetylase, beta/alpha-barrel; Polysaccharide deacetylase | |
| 2574999 | -2.006 | catalytic activity, protein kinase activity, ATP binding | protein amino acid phosphorylation | Six-hairpin glycosidase-like; Protein kinase ATP binding, conserved site | |
| 2740159 | -2.015 | carbohydrate binding, hydrolase activity, hydrolyzing O-glycosyl compounds | carbohydrate metabolism | Carbohydrate-binding family V/XII | |
| 2586510 | -2.020 | hydrolase activity, acting on carbon-nitrogen (but not peptide) bonds | | Metal-dependent hydrolase, composite | |
| 2681191 | -2.025 | catalytic activity, hydrolase activity, acting on carbon-nitrogen (but not peptide) bonds, hydrolase activity | metabolism | Haloacid dehalogenase-like hydrolase; Metal-dependent hydrolase, composite; Amidohydrolase 1 | |
| 2572767 | -2.033 | oxidoreductase activity | metabolism | Aromatic-ring hydroxylase | |
| 2452507 | -2.064 | | | | |
| 2592701 | -2.119 | | | | |
| 2680985 | -2.157 | catalytic activity, FAD binding, oxidoreductase activity | | FAD-binding, type 2 | |
| 2753486 | -2.164 | | | | |
| 2556613 | -2.182 | | | | |
| 2460595 | -2.193 | | | | |
| 2597843 | -2.206 | | | | |
| 2597620 | -2.213 | | | | |
| 2075664 | -2.217 | response to pH, proteolysis and peptidolysis | | Signal transduction response regulator, pH-responsive, PalI/Rim9 | |
| 2678063 | -2.221 | catalytic activity, binding, coenzyme binding | metabolism | NAD-dependent epimerase/dehydratase | |
| 2472024 | -2.304 | response to pH, proteolysis and peptidolysis | | Signal transduction response regulator, pH-responsive, PalI/Rim9 | |
| 2471655 | -2.328 | | | | |
| 2677242 | -2.329 | hydrolase activity, acting on carbon-nitrogen (but not peptide) bonds, hydrolase activity | | Metal-dependent hydrolase, composite; Amidohydrolase 1 | |
| 2117151 | -2.356 | | | | |
| 2702456 | -2.389 | protein kinase activity, protein-tyrosine kinase activity, protein serine/threonine kinase activity, ATP binding | protein amino acid phosphorylation | Protein kinase; Serine/threonine protein kinase-related | |
| 2463002 | -2.408 | | | | |
| 2703675 | -2.412 | aspartic-type endopeptidase activity, pepsin A activity | proteolysis and peptidolysis | Peptidase aspartic | |
| 2283081 | -2.481 | | metabolism | Aminotransferase, class V/Cysteine desulphurase | |
| 2678282 | -2.490 | two-component sensor molecule activity G-protein coupled photoreceptor activity, protein-histidine kinase activity, two-component | regulation of transcription, peptidyl-histidine phosphorylation, sensory perception, | Signal transduction histidine kinase; Phytochrome chromophore attachment domain | |

| | | | | | |
|---|---|---|---|---|---|
| | | response regulator activity, signal transducer activity, ATP binding, transferase activity, transferring phosphorus-containing groups | protein-chromophore linkage, signal transduction, phosphorylation, two-component signal transduction system (phosphorelay), regulation of transcription, DNA-dependent | | |
| 2600031 | -2.494 | hydrolase activity, hydrolyzing O-glycosyl compounds | carbohydrate metabolism | Glycoside hydrolase, family 5 | |
| 2580701 | -2.513 | two-component sensor molecule activity, protein-histidine kinase activity, two-component response regulator activity, signal transducer activity, ATP binding, transferase activity, transferring phosphorus-containing groups | signal transduction, phosphorylation, regulation of transcription, DNA-dependent, two-component signal transduction system (phosphorelay), peptidyl-histidine phosphorylation | Signal transduction histidine kinase | |
| 2621084 | -2.545 | | | | |
| 2696916 | -2.582 | O-methyltransferase activity | | O-methyltransferase, family 2 | |
| 2264463 | -2.604 | hydrolase activity, hydrolyzing O-glycosyl compounds | carbohydrate metabolism | Glycoside hydrolase, family 17 | |
| 2532725 | -2.661 | oxidoreductase activity, acting on paired donors, with oxidation of a pair of donors resulting in the reduction of molecular oxygen to two molecules of water, oxidoreductase activity | lipid metabolism | Fatty acid desaturase, type 1 | |
| 2567631 | -2.740 | | | | Information storage and processing |
| 2708788 | -2.777 | protein kinase activity, protein-tyrosine kinase activity, protein serine/threonine kinase activity, ATP binding | protein amino acid phosphorylation | Protein kinase; Serine/threonine protein kinase-related; Protein kinase ATP binding, conserved site | |
| 2608339 | -2.827 | | | | |
| 2639294 | -2.830 | protein modification | | Ubiquitin | |
| 2530685 | -2.868 | hydrolase activity, acting on ester bonds | lipid metabolism | Lipase, GDSL | |
| 2664870 | -2.967 | hydrogen ion transporter activity, O-methyltransferase activity, zinc ion binding, DNA binding, transcription factor activity | regulation of transcription, DNA-dependent, transcription | ATPase, F1/V1/A1 complex, alpha/beta subunit, nucleotide-binding; O-methyltransferase, family 3; Fungal specific transcription factor | Metabolism |
| 2535816 | -3.010 | | | | |
| 2221890 | -3.084 | catalytic activity, carbon-carbon lyase activity | aromatic compound metabolism | Pyruvate/Phosphoenolpyruvate kinase, catalytic core; HpcH/HpaI aldolase | |

| | | | | | |
|---|---|---|---|---|---|
| 2539960 | -3.093 | hydrolase activity, hydrolyzing O-glycosyl compounds | carbohydrate metabolism | Glycoside hydrolase, family 3 | |
| 2508802 | -3.114 | | | | |
| 2184213 | -3.123 | | | | |
| 2595350 | -3.159 | GTP binding | protein transport, small GTPase mediated signal transduction | Ras small GTPase, Ras type; Ras small GTPase, Rho type; Ras small GTPase, Rab type; Small GTP-binding protein | |
| 2597062 | -3.173 | mannose-1-phosphate guanylyltransferase (GDP) activity, nucleotidyltransferase activity | polysaccharide metabolism, biosynthesis | Mannose-6-phosphate isomerase, type III; Nucleotidyl transferase | |
| 2678193 | -3.186 | monooxygenase activity, oxidoreductase activity | electron transport, aromatic compound metabolism, metabolism | Monooxygenase, FAD-binding; Aromatic-ring hydroxylase | |
| 2678455 | -3.243 | | | | |
| 2528625 | -3.258 | | | | |
| 2555894 | -3.269 | | | | |
| 2619664 | -3.402 | | | | |
| 2746951 | -3.453 | | | | |
| 2546907 | -3.568 | carbohydrate binding, catalytic activity, glucan 1,4-alpha-glucosidase activity | polysaccharide catabolism, polysaccharide metabolism, carbohydrate metabolism | Carbohydrate-binding-like fold; Glycoside hydrolase, carbohydrate-binding; Six-hairpin glycosidase-like | |
| 2439795 | -3.608 | | | | |
| 2672812 | -3.639 | | | | |
| 2198994 | -3.861 | | | | |
| 2677399 | -4.003 | catalytic activity, hydrolase activity, acting on ester bonds | metabolism | Alkaline-phosphatase-like, core domain; Phosphoesterase | |
| 2606067 | -4.203 | | | | Cellular processes and signaling |
| 2548110 | -4.292 | | | | |
| 2540154 | -4.409 | | | | |
| 2195418 | -4.534 | | | | |
| 2689252 | -4.542 | nucleotide, binding, tRNA ligase, ATP binding, oxidoreductase activity | amino acid activation, protein biosynthesis | Aminoacyl-tRNA synthetase, class I, conserved site | |
| 2640541 | -4.610 | | | | |
| 2594802 | -4.659 | | | | |
| 2674326 | -4.917 | microtubule motor activity, ATP binding | microtubule-based movement | Kinesin, motor region | |
| 2602110 | -4.990 | | | | |
| 2574569 | -5.475 | | | | |
| 2680457 | -5.545 | zinc ion binding | | Zinc finger, MYND-type | |
| 2599803 | -5.601 | glutamate decarboxylase, carboxy-lyase activity, transcription factor activity, pyridoxal phosphate binding | carboxylic acid metabolism, glutamate metabolism, regulation of transcription, DNA-dependent | Pyridoxal phosphate-dependent decarboxylase; Glutamate decarboxylase; Homeobox | |

| | | | | | |
|---|---|---|---|---|---|
| 2460130 | -5.695 | | | | |
| 2599368 | -5.809 | | | Peptidase S54, rhomboid | |
| 2537084 | -5.967 | | | | |
| 2742326 | -6.731 | | | | |
| 1138359 | -7.985 | | | | |

### Table S18. Repressed proteins in primordia vs. monokaryon 4-39

| Schco3 ID | Fold change | Go Desc molecular function | Go Desc Biological Process | Interpro Desc | KOG Group |
|---|---|---|---|---|---|
| 2466266 | -2.030 | | | | |
| 2678178 | -2.011 | catalytic activity, aspartic-type endopeptidase activity, pepsin A activity | metabolism, proteolysis and peptidolysis | Crotonase; Peptidase A1 | |
| 2545132 | -2.034 | GTPase activity, structural molecule activity, GTP binding | microtubule-based movement, microtubule-based process | Beta tubulin | |
| 2498761 | -2.051 | | | | |
| 2227218 | -2.121 | nucleoside triphosphatase activity, nucleotide binding, hydrolase activity, ATP binding | protein catabolism | AAA+ ATPase, core; 26S proteasome subunit P45 | |
| 2482758 | -2.128 | nucleoside triphosphatase activity, nucleotide binding, hydrolase activity, ATP binding | protein catabolism | AAA+ ATPase, core; 26S proteasome subunit P45 | |
| 2744020 | -2.132 | protein binding, GTP binding, GTPase activity | nucleocytoplasmic transport, signal transduction, small GTPase mediated signal transduction, protein transport, intracellular protein transport | Ras small GTPase, Ras type; Ras small GTPase, Rho type; Ras small GTPase, Rab type; Ran GTPase; Small GTP-binding protein | |
| 2271670 | -2.146 | endopeptidase activity, threonine endopeptidase activity | ubiquitin-dependent protein catabolism | 20S proteasome, A and B subunits; Proteasome alpha-subunit, conserved site | |
| 2723169 | -2.161 | protein transporter activity, binding | protein-nucleus import, docking, intracellular protein transport | Armadillo-type fold; Importin-beta | |
| 2378688 | -2.265 | | | | |
| 2678491 | -2.269 | guanyl-nucleotide exchange factor activity, isomerase activity, small GTPase regulatory/interacting protein activity, Rho guanyl-nucleotide exchange factor activity | intracellular signaling cascade, cell redox homeostasis, regulation of Rho protein signal transduction | Guanine-nucleotide dissociation stimulator, CDC24, conserved site; Disulphide isomerase; Citron-like | |
| 2531252 | -2.402 | UDP-glucose 4-epimerase activity, catalytic activity, binding, coenzyme binding | metabolism, galactose metabolism | UDP-glucose 4-epimerase | |

| | | | | | |
|---|---|---|---|---|---|
| 2600989 | -2.559 | ATP binding, binding, DNA binding, DNA topoisomerase type II activity | signal transduction, DNA metabolism, transport, DNA topological change | Spo11/DNA topoisomerase VI, subunit A; Mitochondrial substrate carrier; RhoGAP; Rho GTPase | |
| 2675738 | -2.612 | intracellular protein transport | | NSF attachment protein | |
| 2208671 | -2.633 | actin binding, calcium ion binding | | Actinin-type, actin-binding, conserved site; Calcium-binding EF-hand | |
| 2596185 | -3.042 | protein binding, GTP binding, GTPase activity | nucleocytoplasmic transport, signal transduction, small GTPase mediated, signal transduction, protein transport, intracellular protein transport | Ras small GTPase, Ras type; Ras small GTPase, Rho type; Ras small GTPase, Rab type; Ran GTPase; Small GTP-binding protein | |
| 2602003 | -3.112 | protein binding, binding, ATP binding, RNA ligase (ATP) activity | tRNA splicing | DEATH-like; Armadillo-type fold; tRNA ligase, phosphodiesterase | |
| 2602808 | -3.113 | protein prenyltransferase activity | protein amino acid prenylation | Protein prenyltransferase, alpha subunit | |
| 2271597 | -3.115 | | intracellular transport | Ran Binding Protein 1 | |
| 2249278 | -3.257 | | | | |
| 2674535 | -3.382 | protein binding, ATP binding | protein folding | Chaperonin Cpn60; T-complex protein 1, alpha subunit | |
| 2595504 | -3.443 | protein kinase activity, protein serine/threonine kinase activity, nucleic acid binding, ATP binding, protein-tyrosine kinase activity, ATPase activity, voltage-dependent ion-selective channel activity | protein amino acid phosphorylation, anion transport | Protein kinase; ABC transporter-like; Serine/threonine protein kinase-related; Tyrosine protein kinase; mitochondrial outer membrane | |
| 2597008 | -3.988 | protein folding | | Heat shock protein DnaJ | |
| 2599607 | -4.021 | glutathione peroxidase activity | response to oxidative stress | Glutathione peroxidase | |
| 2678444 | -4.653 | protein kinase activity, protein-tyrosine kinase activity, protein serine/threonine kinase activity, ATP binding, N-acetyltransferase activity | protein amino acid phosphorylation, metabolism | Protein kinase; Serine/threonine protein kinase-related; Tyrosine protein kinase | |
| 2490330 | -4.691 | | | | |
| 2548110 | -5.636 | chaperone activity | protein folding | Heat shock protein DnaJ | |
| 2680097 | -6.292 | acid phosphatase activity, zinc ion binding, transcription factor activity, ARF guanyl-nucleotide exchange factor activity | regulation of transcription, DNA-dependent | Histidine acid phosphatase; Zinc finger, C2H2-type; SEC7-like | |
| 2699882 | -6.388 | nucleic acid binding, peptidyl-prolyl cis-trans isomerase activity | protein folding | RNA recognition motif, RNP-1; Peptidyl-prolyl cis-trans isomerase, cyclophilin-type | |
| 2677294 | -7.804 | catalytic activity, binding, coenzyme binding | metabolism | NAD-dependent epimerase/dehydratase | |
| 2678425 | -2.009 | protein binding, GTP binding, GTPase activity | nucleocytoplasmic transport, | Ras small GTPase, Ras type; Ras small GTPase, Rho type; Ras small | |

| | | | | | |
|---|---|---|---|---|---|
| | | | signal transduction, DNA repair, small GTPase mediated signal transduction, protein transport, intracellular protein transport | GTPase, Rab type; Ran GTPase; Small GTP-binding protein | |
| 2166139 | -2.052 | structural constituent of ribosome | protein biosynthesis | Ribosomal protein S9 | |
| 2674545 | -2.053 | structural constituent of ribosome | protein biosynthesis | Ribosomal protein S2 | |
| 2602338 | -2.265 | tRNA-pseudouridine synthase I activity, nucleic acid binding, ATP binding, DNA binding, helicase activity, tRNA binding, ATPase activity | pseudouridine synthesis, tRNA processing | SNF2-related; DNA/RNA helicase; ABC transporter-like | |
| 2147637 | -2.319 | aspartate-tRNA ligase activity, nucleic acid binding, ATP binding, nucleotide binding, transporter activity, tRNA ligase activity | amino acid activation, protein biosynthesis, aspartyl-tRNA aminoacylation, transport | Aminoacyl-tRNA synthetase, class II (D, K and N); Aspartyl-tRNA synthetase, class II | |
| 2743097 | -2.388 | catalytic activity, binding | carbohydrate metabolism | Glycosyl hydrolase, family 13, subfamily, catalytic region; Armadillo-type fold | |
| 2698255 | -2.419 | structural constituent of ribosome | protein biosynthesis | Ribosomal protein L22/L17 | |
| 1319169 | -2.563 | transition metal ion binding, heme binding, nucleic acid binding, zinc ion binding | | Cytochrome b5 | |
| 2676065 | -2.677 | protein methyltransferase activity, iron ion binding, oxidoreductase activity | protein amino acid methylation | Ribosomal L11 methyltransferase; Isopenicillin N synthase | |
| 2746016 | -2.747 | protein binding | | Leucine-rich repeat | |
| 2598258 | -2.777 | carbon-nitrogen ligase activity, with glutamine as amido-N-donor, binding | | Amidase signature enzyme; Armadillo-type fold | |
| 2488899 | -2.825 | structural constituent of ribosome | protein biosynthesis | Ribosomal protein S27e | |
| 2491982 | -2.873 | structural constituent of ribosome, RNA binding | protein biosynthesis | Ribosomal protein L23, conserved site | |
| 2451194 | -2.978 | DNA binding, zinc ion binding | | SANT, DNA-binding; Zinc finger ZZ type | |
| 2596995 | -3.011 | nucleic acid binding, ATP binding, DNA binding, helicase activity, hydrolase activity, hydrolyzing O-glycosyl compounds | carbohydrate metabolism | DNA/RNA helicase; SNF2-related | |
| 2678650 | -3.046 | structural constituent of ribosome | protein biosynthesis | Ribosomal protein L31e | |
| 2677971 | -3.073 | GTPase activity, GTP binding | | Protein synthesis factor, GTP-binding; Translation elongation factor EFTu/EF1A, domain 2 | |
| 2734789 | -3.176 | | | | |
| 2620095 | -3.313 | | | | |
| 2696887 | -3.395 | protein binding, DNA binding, transcription factor activity, oxidoreductase activity | electron transport, regulation of | WW/Rsp5/WWP; Histone-fold; FMN-dependent alpha-hydroxy acid dehydrogenase; Bacterial regulatory protein, LuxR | |

| | | | | | |
|---|---|---|---|---|---|
| | | | transcription, DNA-dependent | | |
| 2749322 | -3.427 | structural constituent of ribosome | protein biosynthesis | Ribosomal protein S17e | |
| 2681298 | -3.433 | thiamin diphosphokinase activity, ATP binding, nucleotide binding, glycine-tRNA, ligase activity, tRNA ligase activity | thiamin diphosphate biosynthesis, amino acid activation, thiamin metabolism, protein biosynthesis, glycyl-tRNA aminoacylation | Thiamin pyrophosphokinase; Glycyl-tRNA synthetase, alpha2 dimer; Aminoacyl-tRNA synthetase, class II (G, H, P and S); Anticodon-binding | |
| 2106720 | -3.606 | ribosome binding | mature ribosome assembly | Translation initiation factor IF6 | |
| 2623763 | -3.606 | | | SET | Information storage and processing |
| 2680491 | -4.186 | protein binding; ATPase activity; ATP binding | DNA metabolism | SMCs flexible hinge; RecF/RecN/SMC protein; ABC transporter-like | |
| 2601895 | -7.587 | calcium ion binding, ATP binding | mismatch repair | Calcium-binding EF-hand; DNA mismatch repair protein; ATP-binding region, ATPase-like | |
| 2274225 | -2.032 | metal ion binding, superoxide dismutase activity | superoxide metabolism | Manganese and iron superoxide dismutase | |
| 2602397 | -2.045 | catalytic activity, malate dehydrogenase activity, binding, oxidoreductase activity, acting on the CH-OH group of donors, NAD or NADP as , L-malate dehydrogenase activity, oxidoreductase activity | glycolysis, carbohydrate metabolism, TCA intermediate metabolism, malate metabolism, metabolism | Lactate dehydrogenase/glycoside hydrolase, family 4; Malate dehydrogenase, active site | |
| 2600373 | -2.055 | oxidoreductase activity, acting on sulfur group of donors, disulfide as acceptor, transcription factor activity | regulation of transcription, DNA-dependent, protein metabolism | Homeobox; Methionine sulphoxide reductase A | |
| 2680146 | -2.059 | nucleic acid binding, ATP binding, nucleotide binding, nucleoside triphosphatase activity, ATPase activity, ATPase activity, coupled to transmembrane movement of substances | transport | RNA recognition motif, RNP-1; ABC transporter-like; ABC transporter-like | |
| 2745344 | -2.068 | protein binding, zinc ion binding, electron carrier activity | electron transport | Electron transfer flavoprotein, beta-subunit, core; Zinc finger, RING-type | |
| 2680115 | -2.081 | catalytic activity | | UBA/THIF-type NAD/FAD binding fold | |
| 2619084 | -2.082 | glycerone kinase activity, ATP binding | glycerol metabolism | Dak kinase; Dak phosphatase | Metabolism |
| 2489815 | -2.096 | catalytic activity, fumarate hydratase activity | fumarate metabolism | Fumarate hydratase, class II; L-Aspartase-like | |
| 2637783 | -2.107 | catalytic activity, carboxy-lyase activity, magnesium ion binding | | Thiamine pyrophosphate-requiring enzyme; Pyruvate decarboxylase/indolepyruvate decarboxylase | Metabolism |

| | | | | | |
|---|---|---|---|---|---|
| 2675304 | -2.140 | beta-galactosidase activity | carbohydrate metabolism | Glycoside hydrolase, family 35 | |
| 2599948 | -2.150 | catalytic activity, binding, ATP binding, oxidoreductase activity | fructose 2,6-bisphosphate metabolism, metabolism | Fructose-2,6-bisphosphatase; Short-chain dehydrogenase/reductase SDR; Glucose/ribitol dehydrogenase | |
| 2276847 | -2.164 | oxidoreductase activity | electron transport | NADH dehydrogenase (ubiquinone), 24 kDa subunit | |
| 2600574 | -2.192 | nucleobase transporter activity, glycogen (starch) synthase activity | glycogen biosynthesis, nucleobase, nucleoside, nucleotide and nucleic acid transport | Permease for cytosine/purines, uracil, thiamine, allantoin; Glycogen synthase | |
| 2597398 | -2.207 | catalytic activity, nucleic acid binding, zinc ion binding, ARF GTPase activator activity, alpha, alpha-trehalase activity | mRNA processing, trehalose metabolism | Six-hairpin glycosidase-like; RNA recognition motif, RNP-1; Arf GTPase activating protein; Splicing factor PWI; Glycoside hydrolase, family 37 | |
| 2032525 | -2.213 | | | | |
| 2679037 | -2.214 | GTP binding, oxidoreductase activity | cell cycle | Cell division/GTP binding protein; Redoxin | |
| 2562064 | -2.227 | phosphoglycerate kinase activity | glycolysis | Phosphoglycerate kinase | |
| 2681810 | -2.263 | hydrolase activity, oxidoreductase activity | | Metallophosphoesterase; Aldo/keto reductase | |
| 2620107 | -2.273 | catalytic activity, oxidoreductase activity, acting on CH-OH group of donors, binding, oxidoreductase activity, acting on the CH-OH group of donors, NAD or NADP as acceptor, glycerol-3-phosphate dehydrogenase (NAD) activity, oxidoreductase activity | glycerol-3-phosphate catabolism, glycerol-3-phosphate metabolism, carbohydrate metabolism, metabolism | NAD-dependent glycerol-3-phosphate dehydrogenase; 6-phosphogluconate dehydrogenase | |
| 2688856 | -2.275 | nucleoside triphosphatase activity, nucleotide binding, ATPase activity, coupled to transmembrane movement of substances, ATPase activity, ATP binding | transport | AAA+ ATPase, core; ABC transporter-like | |
| 2003623 | -2.304 | ribose-phosphate diphosphokinase activity, magnesium ion binding, transferase activity | ribonucleoside monophosphate biosynthesis, nucleotide biosynthesis, nucleoside metabolism | Phosphoribosyl pyrophosphokinase | |
| 2679601 | -2.339 | catalytic activity, cofactor binding, oxidoreductase activity, acting on the CH-OH group of donors | metabolism | D-isomer specific 2-hydroxyacid dehydrogenase, NAD-binding | |
| 2338336 | -2.348 | catalytic activity | metabolism | Isocitrate lyase and phosphorylmutase; Pyruvate/Phosphoenolpyruvate kinase, catalytic core | |
| 2703762 | -2.349 | protein kinase activity, protein serine/threonine kinase activity, ATP binding | actin filament polymerization, protein amino acid phosphorylation | Protein kinase; ARP23 complex 20 kDa subunit | |
| 2282069 | -2.358 | oxidoreductase activity | metabolism | MaoC-like dehydratase | |

| | | | | | |
|---|---|---|---|---|---|
| 2295738 | -2.462 | metal ion transporter activity, zinc ion transporter activity | zinc ion transport, metal ion transport | Zinc/iron permease | |
| 2562773 | -2.474 | beta-N-acetylhexosaminidase activity | carbohydrate metabolism | Glycoside hydrolase, family 20 | |
| 2508662 | -2.502 | cysteine-type endopeptidase activity | proteolysis and peptidolysis | Peptidase C1B, bleomycin hydrolase | |
| 2511932 | -2.509 | catalytic activity, binding, oxidoreductase activity | electron transport, metabolism | Oxidoreductase | |
| 2255769 | -2.512 | | | Translationally controlled tumour-associated TCTP | |
| 2674424 | -2.558 | catalytic activity, binding, oxidoreductase activity | metabolism | Short-chain dehydrogenase/reductase SDR; Glucose/ribitol dehydrogenase | |
| 2750135 | -2.581 | glucosamine-6-phosphate deaminase activity | N-acetylglucosamine metabolism, carbohydrate metabolism | Glucosamine-6-phosphate isomerase | |
| 2620653 | -2.638 | binding | transport | Mitochondrial substrate carrier | |
| 2595597 | -2.656 | protein kinase activity, protein-tyrosine kinase activity, protein serine/threonine kinase activity, ATP binding | protein amino acid phosphorylation | Protein kinase; Serine/threonine protein kinase-related; Tyrosine protein kinase | |
| 2679415 | -2.713 | beta-galactosidase activity | carbohydrate metabolism | Glycoside hydrolase, family 35 | |
| 2602081 | -2.723 | catalytic activity, binding | metabolism | NAD(P)-binding | |
| 2489166 | -2.739 | protein binding, GTP binding | cell cycle | Septin; Cell division/GTP binding protein | |
| 2599939 | -2.761 | catalytic activity, magnesium ion binding | | Thiamine pyrophosphate enzyme, central region | |
| 2273629 | -2.866 | protein binding | cell communication | Phox-like | |
| 2599143 | -2.882 | catalytic activity | metabolism | Thiolase-like | |
| 2591856 | -2.906 | transporter activity | transport | Major facilitator superfamily | |
| 2530104 | -2.920 | oxidoreductase activity | | NADH:flavin oxidoreductase/NADH oxidase | |
| 2274136 | -2.926 | | | | |
| 2675904 | -2.986 | hydrolase activity, hydrolyzing O-glycosyl compounds, oxidoreductase activity | carbohydrate metabolism | Glycoside hydrolase family 2, immunoglobulin-like beta-sandwich; Aldo/keto reductase | |
| 2455051 | -3.090 | catalytic activity | metabolism | Thiolase-like | |
| 2742382 | -3.179 | | copper ion binding, oxidoreductase activity | Multicopper oxidase, type 2 | |
| 2697037 | -3.221 | catalytic activity, cofactor binding, oxidoreductase activity, acting on the CH-OH group of donors, NAD or NADP as acceptor, binding | metabolism | D-isomer specific 2-hydroxyacid dehydrogenase, NAD-binding | |
| 2535945 | -3.278 | phosphoribosylaminoimidazolesuccinocarboxamide synthase activity | purine nucleotide biosynthesis | SAICAR synthetase | |
| 2666754 | -3.463 | glutamate-ammonia ligase activity | glutamine biosynthesis, nitrogen metabolism | Glutamine synthetase, beta-Grasp | |
| 2645499 | -3.620 | | | | |

| | | | | | |
|---|---|---|---|---|---|
| 2470775 | -3.662 | serine carboxypeptidase activity | proteolysis and peptidolysis | Peptidase S10, serine carboxypeptidase | |
| 2676596 | -3.670 | catalytic activity, carboxyl- and arbamoyltransferase activity, monooxygenase activity, binding, amino acid binding, oxidoreductase activity | electron transport, amino acid metabolism, aromatic compound metabolism metabolism | Aspartate/ornithine carbamoyltransferase; Monooxygenase, FAD-binding; Glucose/ribitol dehydrogenase; Pyridine nucleotide-disulphide oxidoreductase, class I; Aromatic-ring hydroxylase; Short-chain dehydrogenase/reductase SDR | |
| 2737131 | -3.747 | FAD binding, oxidoreductase activity | electron transport | Pyridine nucleotide-disulphide oxidoreductase, class I | |
| 2623304 | -3.806 | chitinase activity, hydrolase activity, hydrolyzing O-glycosyl compounds | chitin catabolism, carbohydrate metabolism | Glycoside hydrolase, family 18, catalytic domain; Chitinase II | |
| 2679035 | -3.923 | protein binding | | BAR | |
| 2583245 | -4.021 | serine carboxypeptidase activity, serine-type peptidase activity | proteolysis and peptidolysis | Peptidase S10, serine carboxypeptidase; Peptidase S26A, signal peptidase I | |
| 2695732 | -4.096 | transferase activity, transferring nitrogenous groups, pyridoxal phosphate binding | biosynthesis | Aminotransferase, class I and II | |
| 2707033 | -4.621 | oxidoreductase activity | | Aldo/keto reductase | |
| 2569226 | -5.042 | oxidoreductase activity | | Aldo/keto reductase | |
| 2546713 | -5.228 | catalytic activity, binding | metabolism | NAD(P)-binding | Metabolism |
| 2546053 | -5.457 | oxidoreductase activity | | Aldo/keto reductase | |
| 2744912 | -6.127 | hydrolase activity, hydrolyzing O-glycosyl compounds | carbohydrate metabolism | Glycoside hydrolase, family 3 | |
| 2598771 | -6.245 | oxidoreductase activity | electron transport | FMN-dependent alpha-hydroxy acid dehydrogenase | |
| 2595248 | -6.513 | serine carboxypeptidase activity | proteolysis and peptidolysis | Peptidase S10, serine carboxypeptidase | |
| 2530747 | -7.447 | | | | |
| 2549492 | -2.056 | hydrolase activity, hydrolyzing O-glycosyl compounds | carbohydrate metabolism | Glycoside hydrolase, family 5 | |
| 2716936 | -2.179 | monooxygenase activity, peroxidase activity, heme binding, iron ion binding | electron transport, response to oxidative stress | Cytochrome P450; Haem peroxidase | |
| 2601059 | -2.215 | | | | |
| 2608284 | -2.231 | carboxypeptidase A activity, chaperone activator activity, ATPase stimulator activity, zinc ion binding | proteolysis and peptidolysis | Peptidase M14, carboxypeptidase A; Activator of Hsp90 ATPase | |
| 2669543 | -2.259 | GTP binding | protein transport, small GTPase mediated signal transduction | Ras small GTPase, Ras type; Ras small GTPase, Rho type; Ras small GTPase, Rab type; Small GTP-binding protein | |
| 2198127 | -2.407 | | | | |
| 2590711 | -2.487 | oxidoreductase activity | | Aldo/keto reductase | |
| 2597780 | -2.559 | catalytic activity, sugar porter activity, protein serine/threonine phosphatase activity, transporter activity | carbohydrate transport, protein amino acid dephosphorylation, transport | Protein phosphatase 2C-related; General substrate transporter; Major facilitator superfamily | |
| 2677972 | -2.567 | structural constituent of ribosome, GTP binding | protein transport, small GTPase | Ribosomal protein S7e; Ras small GTPase, Ras type; Ras small GTPase, Rho type; Ras small | |

| | | | | | |
|---|---|---|---|---|---|
| | | | mediated signal transduction, protein biosynthesis | GTPase, Rab type; Small GTP-binding protein | |
| 2750138 | -2.626 | FAD binding, oxidoreductase activity, acting on CH-OH group of donors | alcohol metabolism | Glucose-methanol-choline oxidoreductase | |
| 2600127 | -2.661 | | | | |
| 2600743 | -2.759 | protein binding, oxidoreductase activity | | BTB/POZ; Aldo/keto reductase | |
| 2598891 | -2.885 | | | | |
| 2615761 | -3.072 | thiamin biosynthesis | | Thiamine biosynthesis Thi4 protein | |
| 2573839 | -3.095 | GTPase activity, GTP binding | | Dynamin, GTPase region | |
| 2675760 | -3.110 | hydrolase activity | | Dienelactone hydrolase | |
| 2548824 | -3.224 | FAD binding, oxidoreductase activity, acting on CH-OH group of donors | alcohol metabolism | Glucose-methanol-choline oxidoreductase | |
| 2695583 | -3.344 | FAD binding, caspase activity, oxidoreductase activity, acting on CH-OH group of donors | proteolysis and peptidolysis | Glucose-methanol-choline oxidoreductase; Peptidase C14, caspase catalytic | |
| 2573733 | -3.454 | FAD binding, oxidoreductase activity, acting on CH-OH group of donors | alcohol metabolism | Glucose-methanol-choline oxidoreductase | |
| 2595836 | -3.577 | catalytic activity, binding, oxidoreductase activity | metabolism | Short-chain dehydrogenase/reductase SDR; Glucose/ribitol dehydrogenase | |
| 2693747 | -3.616 | transporter activity, binding | signal transduction, transport | Lipocalin; Rho GTPase activation protein | |
| 2599947 | -3.795 | | | | |
| 2502834 | -4.185 | hydrolase activity | | Dienelactone hydrolase | |
| 2688236 | -4.394 | catalytic activity, binding, ATP binding, nucleotide binding, tRNA ligase activity, oxidoreductase activity | amino acid activation, protein biosynthesis, metabolism | Aminoacyl-tRNA synthetase, class I, conserved site; Short-chain dehydrogenase/reductase SDR; Glucose/ribitol dehydrogenase | |
| 2480821 | -4.881 | nucleic acid binding | RNA recognition motif, RNP-1 | | |
| 2485740 | -5.245 | | | | |
| 2660812 | -5.957 | oxidoreductase activity | negative regulation of transcription | Flavodoxin/nitric oxide synthase; Flavoprotein WrbA | |
| 2693329 | -2.019 | | | | |
| 2703675 | -2.040 | aspartic-type endopeptidase activity, pepsin A activity | proteolysis and peptidolysis | Peptidase aspartic, catalytic | |
| 2701600 | -2.082 | cysteine-type endopeptidase activity, nutrient reservoir activity | proteolysis and peptidolysis | Peptidase, cysteine peptidase active site | |
| 2580789 | -2.096 | | | | |
| 2667616 | -2.113 | nucleic acid binding, exonuclease activity | | Putative 5-3 exonuclease | |
| 2679667 | -2.122 | acid phosphatase activity, nucleic acid binding, zinc ion binding | | Histidine acid phosphatase; Zinc finger, C2H2-type | |
| 2538728 | -2.162 | ribonuclease activity, RNA binding | | Ribonuclease II and R | |
| 2597428 | -2.228 | catalytic activity, binding | metabolism | NAD(P)-binding | |

| | | | | | |
|---|---|---|---|---|---|
| 2702164 | -2.231 | cysteine-type endopeptidase activity, manganese ion binding, pyrophosphatase activity, hydrolase activity | proteolysis and peptidolysis | Peptidase, cysteine peptidase active site; Phosphoesterase, RecJ-like; DHHA2 | |
| 2596872 | -2.235 | | | | |
| 2480617 | -2.242 | transcription factor activity, signal transducer activity | signal transduction, regulation of transcription, DNA-dependent | STAT transcription factor, coiled coil | |
| 2608058 | -2.269 | binding | | Armadillo-type fold | |
| 2512880 | -2.271 | | | | |
| 2493604 | -2.312 | | | | |
| 2598403 | -2.325 | catalytic activity | | Six-hairpin glycosidase-like | |
| 2601175 | -2.364 | zinc ion binding, pepsin A activity, DNA binding, aspartic-type endopeptidase activity, transcription factor activity | regulation of transcription, DNA-dependent, proteolysis and peptidolysis, transcription | Fungal specific transcription factor; Peptidase A1 | |
| 2642225 | -2.413 | | | | |
| 2257329 | -2.452 | oxidoreductase activity | potassium ion transport | Aldo/keto-reductase; Potassium channel, voltage-dependent, beta subunit, KCNAB-related | |
| 2301961 | -2.455 | voltage-gated potassium channel activity | potassium ion transport | KCNAB voltage-gated K+ channel, beta subunit | Metabolism |
| 2603182 | -2.458 | galactokinase activity, phosphotransferase activity, alcohol group as acceptor, ATP binding, kinase activity | carbohydrate phosphorylation, phosphorylation, metabolism, galactose metabolism | Mevalonate and galactokinase; GHMP kinase | |
| 2666791 | -2.467 | serine-type peptidase activity | proteolysis and peptidolysis | Peptidase S28 | |
| 2678063 | -2.474 | catalytic activity, binding, coenzyme binding | metabolism | NAD-dependent epimerase/dehydratase | |
| 2565384 | -2.524 | catalytic activity, binding, oxidoreductase activity | metabolism | 6-phosphogluconate dehydrogenase | |
| 2599368 | -2.532 | | | Peptidase S54, rhomboid | |
| 2513636 | -2.534 | | | | |
| 2580701 | -2.558 | two-component sensor molecule activity, protein-histidine kinase activity, two-component response regulator activity, signal transducer activity, ATP binding, transferase activity, transferring phosphorus-containing groups | signal transduction, phosphorylation, regulation of transcription, DNA-dependent, two-component signal transduction system (phosphorelay), peptidyl-histidine phosphorylation | Signal transduction histidine kinase, core | |
| 2592036 | -2.581 | acid phosphatase activity | | Histidine acid phosphatase | |
| 2599114 | -2.608 | rRNA processing | | Small-subunit processome, Utp13 | |
| 2600484 | -2.637 | | | | |
| 2692586 | -2.678 | hydrolase activity, hydrolyzing O-glycosyl compounds | carbohydrate metabolism | Glycoside hydrolase, family 3 | |

| | | | | | |
|---|---|---|---|---|---|
| 2689658 | -2.708 | | | | |
| 2586510 | -2.723 | hydrolase activity, acting on carbon-nitrogen (but not peptide) bonds | | Metal-dependent hydrolase, composite | |
| 2283081 | -2.773 | | metabolism | Aminotransferase, class V/Cysteine desulphurase | |
| 2600338 | -2.784 | | | | |
| 2699394 | -2.826 | | | | |
| 2508404 | -2.894 | | | | |
| 2594802 | -2.938 | | | | |
| 2708788 | -2.945 | protein kinase activity, protein-tyrosine kinase activity, protein serine/threonine kinase activity, ATP binding | protein amino acid phosphorylation | Protein kinase; Serine/threonine protein kinase-related; Tyrosine protein kinase | |
| 2673553 | -2.957 | catalytic activity, hydrolase activity, acting on carbon-nitrogen (but not peptide) bonds | carbohydrate metabolism | Glycoside hydrolase/deacetylase, beta/alpha-barrel; Polysaccharide deacetylase | |
| 2679088 | -2.974 | hydrolase activity, hydrolyzing O-glycosyl compounds | carbohydrate metabolism | Glycoside hydrolase, family 16 | |
| 2597062 | -3.033 | mannose-1-phosphate guanylyltransferase (GDP) activity, nucleotidyltransferase activity | polysaccharide metabolism, biosynthesis | Mannose-6-phosphate isomerase, type II; Nucleotidyl transferase | |
| 2675318 | -3.150 | | | | |
| 2535816 | -3.193 | | | | |
| 2600930 | -3.198 | catalytic activity, binding, oxidoreductase activity | metabolism | Short-chain dehydrogenase/reductase SDR; Glucose/ribitol dehydrogenase; Aromatic-ring hydroxylase | |
| 2536636 | -3.205 | | | | |
| 2749954 | -3.286 | structural constituent of ribosome | protein biosynthesis | Ribosomal protein S11 | |
| 2734322 | -3.305 | | | | |
| 2589858 | -3.402 | hydrolase activity, hydrolyzing O-glycosyl compounds | carbohydrate metabolism | Glycoside hydrolase, family 6 | |
| 2555894 | -3.471 | | | | |
| 2508802 | -3.477 | | | | |
| 2528310 | -3.486 | | | | |
| 2678455 | -3.521 | | | | |
| 2595350 | -3.532 | GTP binding | protein transport, small GTPase mediated signal transduction | Ras small GTPase, Ras type; Ras small GTPase, Rho type; Ras small GTPase, Rab type; Small GTP-binding protein | |
| 2603513 | -3.703 | catalytic activity, structural constituent of ribosome, binding, transporter activity | protein biosynthesis, transport, metabolism | Ribosomal protein L1; Major intrinsic protein | |
| 2439795 | -3.710 | | | | |
| 2674326 | -3.711 | microtubule motor activity, ATP binding | microtubule-based movement | Kinesin, motor region | |
| 2743762 | -3.732 | racemase and epimerase activity, acting on amino acids and derivatives | metabolism | Asp/Glu racemase | |
| 2539960 | -3.734 | hydrolase activity, hydrolyzing O-glycosyl compounds | carbohydrate metabolism | Glycoside hydrolase, family 3, N-terminal | |

| | | | | | |
|---|---|---|---|---|---|
| 2498164 | -3.738 | hydrolase activity | | Alpha/beta hydrolase | |
| 2117151 | -3.819 | | | | |
| 2574999 | -4.074 | catalytic activity, protein kinase activity, ATP binding | protein amino acid phosphorylation | Six-hairpin glycosidase-like; Protein kinase ATP binding, conserved site | |
| 2740159 | -4.081 | carbohydrate binding, hydrolase activity, hydrolyzing O-glycosyl compounds | carbohydrate metabolism | Carbohydrate-binding family V/XII | |
| 2540154 | -4.115 | | | | |
| 2697931 | -4.120 | | | | |
| 2742326 | -4.212 | | | | |
| 2586789 | -4.237 | | | | |
| 2738673 | -4.304 | GTP binding | protein transport, small GTPase mediated signal transduction | Ras small GTPase, Ras type; Ras small GTPase, Rho type; Ras small GTPase, Rab type; Small GTP-binding protein | |
| 2264463 | -4.599 | hydrolase activity, hydrolyzing O-glycosyl compounds | carbohydrate metabolism | Glycoside hydrolase, family 17 | |
| 2692813 | -4.667 | catalytic activity | metabolism | Mandelate racemase/muconate lactonizing enzyme | |
| 2696064 | -4.690 | FAD binding, oxidoreductase activity, acting on CH-OH group of donors | alcohol metabolism | Glucose-methanol-choline oxidoreductase | |
| 2574569 | -4.710 | | | | |
| 1138359 | -5.042 | | | | |
| 2672812 | -5.189 | | | | |
| 2452507 | -5.322 | | | | |
| 2195418 | -5.386 | | | | |
| 2630944 | -5.506 | | | | |
| 2572813 | -5.570 | protein folding | | Heat shock protein DnaJ | |
| 2680457 | -5.701 | zinc ion binding | | Zinc finger, MYND-type | |
| 2537084 | -6.111 | | | | |
| 1209921 | -6.464 | | | | |
| 2640541 | -6.687 | | | | |
| 2599803 | -6.720 | glutamate decarboxylase activity, carboxy-lyase activity, transcription factor activity, pyridoxal phosphate binding | carboxylic acid metabolism, glutamate metabolism, regulation of transcription, DNA-dependent | Glutamate decarboxylase; Pyridoxal phosphate-dependent decarboxylase; Homeobox | |

**Table S19. Induced proteins in fruiting bodies vs. monokaryon 12-43**

| Schco3 ID | Fold change | Go Desc molecular function | Go Desc Biological Process | Interpro Desc | KOG Group |
|---|---|---|---|---|---|
| 2660823 | 7.901 | metalloendopeptidase activity | proteolysis and peptidolysis | Peptidase M35, deuterolysin | |
| 2679537 | 5.974 | hydrolase activity, hydrolyzing O-glycosyl compounds, RNA binding, nucleic acid binding | RNA processing, carbohydrate metabolism | Glycoside hydrolase; SWAP/Surp; RNA recognition motif | |
| 2549492 | 5.311 | hydrolase activity, hydrolyzing O-glycosyl compounds | carbohydrate metabolism | Glycoside hydrolase, family 5 | |

| | | | | | |
|---|---|---|---|---|---|
| 2587232 | 4.766 | sugar porter activity, transporter activity | carbohydrate transport, transport | General substrate transporter, Major facilitator superfamily, Sugar transporter | |
| 2611041 | 4.653 | | | | Poorly characterized |
| 2695583 | 4.237 | FAD binding, caspase activity, oxidoreductase activity, acting on CH-OH group of donors | proteolysis and peptidolysis | Glucose-methanol-choline oxidoreductase; Peptidase C14, caspase catalytic | |
| 2274818 | 3.616 | nucleic acid binding | | RNA recognition motif, RNP-1 | |
| 2661958 | 3.593 | GTPase activity, GTP binding | | Dynamin, GTPase region | |
| 2695463 | 3.554 | | | | |
| 2110112 | 3.541 | | | Auxin efflux carrier | |
| 2195169 | 3.469 | | | | |
| 2598115 | 3.026 | transporter activity | transport | Major facilitator superfamily | |
| 76079 | 2.985 | sugar porter activity, transporter activity | carbohydrate transport, transport | General substrate transporter; Major facilitator superfamily; Sugar transporter, conserved site | |
| 2597188 | 2.510 | endopeptidase activity, threonine endopeptidase activity, inositol/phosphatidylinositol phosphatase activity | ubiquitin-dependent protein catabolism, protein folding | Proteasome alpha-subunit, conserved site; 20S proteasome, A and B subunits; Inositol monophosphatase | |
| 2674161 | 2.388 | transition metal ion binding, hydrolase activity, oxidoreductase activity | | Ferritin/ribonucleotide reductase-like; Beta lacmatase-like | |
| 2592328 | 2.323 | FAD binding, oxidoreductase activity, acting on CH-OH group of donors | alcohol metabolism | Glucose-methanol-choline oxidoreductase | |
| 2540406 | 2.091 | oxidoreductase activity, acting on the CH-CH group of donors, acyl-CoA dehydrogenase activity | electron transport, metabolism | Acyl-CoA oxidase/dehydrogenase | |
| 2611195 | 2.049 | | | | |
| 2750840 | 8.418 | | | | |
| 2466833 | 8.178 | catalytic activity, FAD binding, oxidoreductase activity | | FAD-binding, type 2; FAD linked oxidase | |
| 2118192 | 7.054 | chitinase activity, hydrolase activity, hydrolyzing O-glycosyl compounds | chitin catabolism, carbohydrate metabolism | Chitinase II; Glycoside hydrolase; | |
| 2625518 | 6.643 | carboxy-lyase activity, pyridoxal phosphate binding | carboxylic acid metabolism | Pyridoxal phosphate-dependent decarboxylase | |
| 2601300 | 6.523 | catalytic activity, binding, zinc ion binding, oxidoreductase activity | metabolism | NAD(P)-binding; Alcohol dehydrogenase; Aldo/keto reductase | |
| 2667549 | 6.490 | monooxygenase activity, heme binding, iron ion binding | electron transport | Cytochrome P450 | |
| 2199166 | 5.939 | amino acid transporter activity | amino acid transport, transport | Amino acid/polyamine transporter I; Amino acid permease-associated region | |
| 2600367 | 5.065 | FAD binding, iron ion binding, oxidoreductase activity | electron transport | Ferric reductase-like transmembrane component | |
| 2680505 | 4.981 | monooxygenase activity, heme binding, iron ion binding | electron transport | Cytochrome P450 | |

| | | | | | |
|---|---|---|---|---|---|
| 48253 | 4.765 | gamma-glutamyltransferase activity | | Gamma-glutamyltranspeptidase | |
| 2472473 | 4.358 | transferase activity, transferring alkyl or aryl (other than methyl) groups, pyridoxal phosphate binding | amino acid metabolism | Cys/Met metabolism, pyridoxal phosphate-dependent enzyme; O-acetylhomoserine/O-acetylserine sulfhydrylase | |
| 2679197 | 4.277 | hydrolase activity, histone deacetylase activity | histone deacetylation | Metallophosphoesterase; Histone deacetylase | |
| 2744134 | 4.247 | transferase activity, transferring hexosyl groups | carbohydrate biosynthesis | Glycosyl transferase, family 8 | |
| 2552181 | 4.141 | oxidoreductase activity | electron transport | Taurine catabolism dioxygenase TauD/TfdA | |
| 2550951 | 3.938 | catalytic activity, binding | metabolism | NAD(P)-binding | |
| 2600607 | 3.929 | | | | |
| 1188538 | 3.847 | arginase activity, hydrolase activity, acting on carbon-nitrogen (but not peptide) bonds, in linear amidines, metal ion binding | arginine catabolism | Arginase; Ureohydrolase | Metabolism |
| 2678898 | 3.717 | guanyl nucleotide binding, carbon-nitrogen ligase activity, with glutamine as amido-N-donor, signal transducer activity | signal transduction, G-protein coupled receptor protein signaling pathway | Guanine nucleotide binding protein (G-protein), alpha subunit; Amidase signature enzyme | |
| 2620653 | 3.629 | binding | transport | Mitochondrial substrate carrier | |
| 2436028 | 3.502 | chorismate synthase activity | aromatic amino acid family biosynthesis | Chorismate synthase | |
| 2596075 | 3.471 | nucleic acid binding, pyridoxal phosphate binding | cysteine metabolism, metabolism | RNA recognition motif, RNP-1; Cysteine desulphurase; Aminotransferase, class V/Cysteine desulphurase | |
| 2674336 | 3.318 | glycolipid 2-alpha-mannosyltransferase activity | protein amino acid glycosylation | Glycosyl transferase, family 15 | |
| 2690581 | 3.137 | catalytic activity, calcium ion binding, alpha-amylase activity | carbohydrate metabolism | Glycosyl hydrolase, family 13, subfamily, catalytic region; Alpha-amylase, fungi | |
| 2679758 | 3.066 | hydrolase activity, hydrolyzing O-glycosyl compounds | carbohydrate metabolism | Glycoside hydrolase family 2, immunoglobulin-like beta-sandwich | |
| 2737131 | 3.051 | FAD binding, oxidoreductase activity | electron transport | Pyridine nucleotide-disulphide oxidoreductase, class I | |
| 2603600 | 2.747 | catalytic activity | cell wall catabolism, metabolism | Dihydroxy-acid and 6-phosphogluconate dehydratase; Peptidoglycan-binding LysM | |
| 2443639 | 2.706 | lactoylglutathione lyase activity | carbohydrate metabolism | Glyoxalase I | |
| 2680579 | 2.616 | catalytic activity, binding, oxidoreductase activity | amino acid metabolism, metabolism | Glutamate/phenylalanine/leucine/valine dehydrogenase | |
| 2597924 | 2.573 | catalytic activity, binding, oxidoreductase activity | metabolism | Flavoprotein; Short-chain dehydrogenase/reductase SDR; Glucose/ribitol dehydrogenase | |
| 2258133 | 2.442 | | metabolism | Aminotransferase, class V/Cysteine desulphurase | |
| 2595827 | 2.291 | catalytic activity, binding, oxidoreductase activity | metabolism | Aldehyde dehydrogenase | |
| 2679876 | 2.262 | catalytic activity, cofactor binding, transporter activity, binding | transport, metabolism | AMP-dependent synthetase and ligase; Phosphopantetheine- | |

| | | | | | |
|---|---|---|---|---|---|
| | | | | binding; Major facilitator superfamily | |
| 2676524 | 2.244 | deaminase activity, AMP deaminase activity | purine ribonucleoside monophosphate biosynthesis | Adenosine/AMP deaminase | |
| 2678866 | 7.530 | | | | |
| 2600178 | 7.465 | beta-catenin binding | development | Beta-catenin-interacting ICAT | |
| 2628500 | 6.727 | | | | |
| 2675600 | 3.914 | hydrolase activity | | NUDIX hydrolase | |
| 2681771 | 3.907 | NADPH binding aspartate-semialdehyde dehydrogenase activity | amino acid metabolism | Aspartate-semialdehyde dehydrogenase, conserved site | |
| 2598981 | 3.488 | zinc ion binding | | Zinc finger, MYND-type; Batten's disease protein Cln3 | |
| 2677822 | 2.840 | ligase activity, ATP binding | mismatch repair | Carboxyl transferase; Acetyl-coenzyme A carboxyltransferase; DNA mismatch repair protein; ATP-binding region, ATPase-like | |
| 2702302 | 2.730 | nucleic acid binding | | Polynucleotidyl transferase, Ribonuclease H fold | |
| 2697922 | 2.675 | nucleic acid binding | | Polynucleotidyl transferase, Ribonuclease H fold | |
| 2676929 | 2.470 | protein binding, DNA binding, zinc ion binding | nucleosome assembly | Zinc finger, RING-type; Histone H3 | |
| 2602443 | 2.373 | nucleic acid binding | | RNA recognition motif, RNP-1 | |
| 2020262 | 2.368 | transferase activity, binding, acyltransferase activity | | Trimeric LpxA-like; Armadillo-type fold | |
| 1319169 | 2.225 | transition metal ion binding, heme binding, nucleic acid binding, zinc ion binding | | Cytochrome b5 | |
| 2603816 | 2.014 | protein binding, binding | metabolism | MIF4G-like, type 2 | |
| 2732602 | 11.862 | peptidase activity | proteolysis and peptidolysis | Peptidase M28 | |
| 2599371 | 8.109 | | | | |
| 2750523 | 7.796 | hydrolase activity | metabolism | Lipase, GDXG, active site | |
| 2353119 | 7.291 | sugar binding, aspartic-type endopeptidase activity, pepsin A activity | proteolysis and peptidolysis | Legume lectin; Peptidase aspartic; Peptidase A1 | |
| 2674662 | 5.407 | microtubule binding | microtubule cytoskeleton organization and biogenesis | HOOK | |
| 2101614 | 5.295 | | | | |
| 2009173 | 5.261 | | | | |
| 2540369 | 5.191 | | | | |
| 2192995 | 4.743 | | | | |
| 2596898 | 3.165 | oxidoreductase activity | metabolism | Aldehyde dehydrogenase, conserved site | |
| 2678033 | 3.061 | | | | |
| 2502077 | 2.863 | | | | |
| 2497887 | 2.528 | peptidase activity | proteolysis and peptidolysis | Peptidase M28 | |
| 2558782 | 2.109 | prolyl oligopeptidase activity, serine-type peptidase activity, serine-type endopeptidase activity | proteolysis and peptidolysis | Peptidase S9A, prolyl oligopeptidase | |
| 2599287 | 2.065 | peptidase activity | proteolysis and peptidolysis | Peptidase M28 | |
| 2618740 | 10.942 | | | | |

| | | | | | |
|---|---|---|---|---|---|
| 2467818 | 10.697 | | | | |
| 2626480 | 10.587 | | | | |
| 2541149 | 10.544 | | | | |
| 2635172 | 10.442 | | | | |
| 2481522 | 10.429 | | | | |
| 2749150 | 9.868 | | | Allergen V5/Tpx-1 related | |
| 2750895 | 9.856 | | | | |
| 2598518 | 9.425 | | | | |
| 2579584 | 9.301 | subtilase activity, serine-type peptidase activity | proteolysis and peptidolysis | Peptidase S8 and S53, subtilisin, kexin, sedolisin | |
| 2602724 | 9.258 | | | | |
| 2470835 | 9.189 | | | | |
| 2541426 | 9.161 | | | | |
| 2308933 | 9.115 | | | Allergen V5/Tpx-1 related | |
| 2493980 | 9.039 | | | | |
| 2295360 | 8.964 | | | | |
| 2701497 | 8.809 | | | | |
| 2749147 | 8.684 | | | Allergen V5/Tpx-1 related | |
| 2703131 | 8.607 | | | | |
| 2549292 | 8.440 | | | | |
| 2678339 | 8.393 | | | | |
| 2745841 | 8.380 | | | | |
| 2534073 | 8.346 | | | | |
| 2636155 | 8.258 | ribonuclease activity, endoribonuclease activity, RNA binding | | Ribonuclease/ribotoxin; Guanine-specific ribonuclease N1 and T1 | |
| 2663658 | 8.148 | | | | |
| 2197213 | 7.649 | | | | |
| 2599691 | 7.609 | | | | |
| 2586726 | 7.597 | | | | |
| 2541924 | 7.447 | | | | |
| 2597881 | 7.446 | aspartic-type endopeptidase activity, pepsin A activity | lipid metabolism, metabolism, proteolysis and peptidolysis | Peptidase aspartic, active site; Peptidase A1; Patatin; Acyl transferase/acyl hydrolase/lysophospholipase | |
| 2696974 | 7.315 | RNA-directed DNA polymerase activity, nucleic, acid binding, zinc ion binding, DNA binding, aspartic-type endopeptidase activity, RNA binding | RNA dependent DNA replication, DNA integration, proteolysis and peptidolysis | RNA-directed DNA polymerase (reverse transcriptase); Zinc finger, CCHC-type; Polynucleotidyl transferase, Ribonuclease H fold; Integrase, catalytic core; Peptidase aspartic, Chromo domain-like | |
| 2350680 | 7.216 | | | | |
| 2503584 | 7.092 | | | | |
| 2442813 | 6.896 | | | | |
| 2570153 | 6.668 | | | | |
| 2638754 | 6.568 | FAD binding, pyranose oxidase activity, oxidoreductase activity, acting on CH-OH group of donors, oxidoreductase activity | glucose metabolism | Glucose-methanol-choline oxidoreductase; Pyranose oxidase; FAD dependent oxidoreductase | |
| 2584950 | 6.483 | | | | |
| 2625719 | 6.376 | aminopeptidase activity, prolyl aminopeptidase activity | proteolysis and peptidolysis | Peptidase S33 | |
| 2701101 | 6.323 | oxidoreductase activity | metabolism | Aldehyde/histidinol dehydrogenase | |
| 1187227 | 6.176 | | | | |

| | | | | | |
|---|---|---|---|---|---|
| 2317702 | 6.066 | nucleic acid binding, zinc ion binding | | Zinc finger | |
| 2677655 | 5.840 | catalytic activity, protein kinase activity, protein serine/threonine kinase activity, binding, ATP binding, protein-tyrosine kinase activity | protein amino acid phosphorylation, metabolism | NAD(P)-binding; Protein kinase; Serine/threonine protein kinase-related; Protein kinase ATP binding; Serine/threonine protein kinase; Tyrosine protein kinase; | |
| 2603514 | 5.655 | nucleic acid binding | | RNA recognition motif | |
| 2468516 | 5.493 | | | | |
| 2602828 | 5.040 | catalytic activity, hydrolase activity, acting on carbon-nitrogen (but not peptide) bonds | carbohydrate metabolism | Glycoside hydrolase/deacetylase, beta/alpha-barrel; Polysaccharide deacetylase | |
| 2642225 | 4.779 | | | | |
| 2581457 | 4.775 | | | | |
| 2667748 | 4.749 | | | | |
| 2743569 | 4.574 | | | | |
| 2748821 | 4.551 | catalytic activity, hydrolase activity, acting on carbon-nitrogen (but not peptide) bonds | carbohydrate metabolism | Glycoside hydrolase/deacetylase, beta/alpha-barrel; Polysaccharide deacetylase | |
| 2600246 | 4.420 | metalloendopeptidase activity, hydrolase activity, hydrolyzing O-glycosyl compounds | carbohydrate metabolism, proteolysis and peptidolysis | Peptidase M35, deuterolysin; Glycoside hydrolase, family 31 | |
| 2545442 | 4.371 | hydrolase activity, hydrolyzing O-glycosyl compounds | carbohydrate metabolism | Glycoside hydrolase, family 3 | |
| 2601114 | 4.277 | transcription factor activity, zinc ion binding | regulation of transcription, DNA-dependent | Fungal transcriptional regulatory protein, N-terminal | |
| 2685946 | 4.174 | oxidoreductase activity | electron transport | Amine oxidase | |
| 2680606 | 4.124 | | | | |
| 2707024 | 4.010 | structural constituent of ribosome | protein biosynthesis | Ribosomal protein L19/L19e | |
| 2535493 | 3.854 | hydrolase activity, hydrolyzing O-glycosyl compounds | carbohydrate metabolism | Glycoside hydrolase, family 5 | |
| 2681665 | 3.650 | | | | |
| 2367013 | 3.545 | | | | |
| 2602331 | 3.472 | | | | |
| 2492670 | 3.368 | | | | |
| 2674206 | 3.286 | nucleic acid binding, iron ion binding, ATP binding, helicase activity, catalase activity, heme binding, RNA binding, ribonuclease III activity, ATP dependent helicase activity | electron transport, RNA processing, response to oxidative stress | DNA/RNA helicase; Catalase; Ribonuclease III | |
| 2639888 | 3.232 | | | | |
| 2529447 | 3.144 | hydrolase activity, hydrolyzing O-glycosyl compounds | carbohydrate metabolism | Glycoside hydrolase, family 5 | |
| 1097623 | 2.915 | | | | |
| 2538638 | 2.809 | catalytic activity, binding, oxidoreductase activity | metabolism | Short-chain dehydrogenase/reductase SDR; Glucose/ribitol dehydrogenase | |
| 2632337 | 2.777 | | | | |
| 2517501 | 2.697 | oxidoreductase activity, acting on single donors with incorporation of | aromatic compound metabolism | Intradiol ring-cleavage dioxygenase, core | |

| | | | | | |
|---|---|---|---|---|---|
| | | molecular oxygen, incorporation of two atoms of oxygen, catalytic activity, ferric iron binding, iron ion binding | | | |
| 2751139 | 2.683 | | | | |
| 2676403 | 2.578 | chorismate mutase activity | aromatic amino acid family biosynthesis | Chorismate mutase; Prohibitin | |
| 2527041 | 2.522 | nucleic acid binding, zinc ion binding | | Zinc finger | |
| 2595539 | 2.432 | | | | |
| 2688935 | 2.398 | | | | |
| 2516775 | 2.330 | | | | |
| 2442811 | 2.329 | | | | |
| 2601373 | 2.321 | | | | |
| 2157839 | 2.272 | | | | |
| 2677004 | 2.198 | | | | |
| 2216136 | 2.180 | structural constituent of ribosome | translational elongation | Ribosomal protein 60S | |
| 2017878 | 2.126 | | | | |
| 1105701 | 2.118 | | | | |

## Table S20. Induced proteins in fruiting bodies vs. monokaryon 4-39

| Schco3 ID | Fold change | Go Desc molecular function | Go Desc Biological Process | Interpro Desc | KOG Group |
|---|---|---|---|---|---|
| 2732602 | 12.862 | peptidase activity | proteolysis and peptidolysis | Peptidase M28 | |
| 2009173 | 10.601 | | | | |
| 2603198 | 9.444 | | | | |
| 2192995 | 8.050 | | | | |
| 2750523 | 7.650 | hydrolase activity | metabolism | Lipase, GDXG, active site | |
| 2674662 | 7.195 | microtubule binding | microtubule cytoskeleton organization and biogenesis | HOOK | |
| 2353119 | 6.831 | sugar binding, aspartic-type endopeptidase activity, pepsin A activity | proteolysis and peptidolysis | Legume lectin, beta domain; Peptidase aspartic, active site; Peptidase A1 | |
| 2497887 | 6.361 | peptidase activity | proteolysis and peptidolysis | Peptidase M28 | |
| 2599371 | 5.912 | | | | |
| 2101614 | 5.705 | | | | |
| 2596898 | 5.598 | oxidoreductase activity | metabolism | Aldehyde dehydrogenase, conserved site | |
| 2540369 | 5.532 | | | | |
| 2678033 | 4.093 | | | | |
| 2596052 | 3.756 | peptidase activity | proteolysis and peptidolysis | Peptidase M28 | |
| 2680703 | 3.427 | glucosylceramidase activity, protein binding, binding | lysosome organization and biogenesis, protein complex assembly, sphingolipid metabolism, intracellular protein transport, vesicle-mediated transport | Glycoside hydrolase, family 30; Clathrin/coatomer adaptor, adaptin-like; Armadillo-type fold; | |

| | | | | | |
|---|---|---|---|---|---|
| 2558782 | 3.396 | prolyl oligopeptidase activity, serine-type peptidase activity, serine-type endopeptidase activity | proteolysis and peptidolysis | Peptidase S9, serine active site | |
| 2599287 | 2.950 | peptidase activity | proteolysis and peptidolysis | Peptidase M28 | |
| 2599027 | 2.942 | membrane dipeptidase activity, dipeptidyl-peptidase activity | proteolysis and peptidolysis | Peptidase M19, renal dipeptidase | |
| 2700135 | 2.602 | | | | |
| 2597644 | 2.511 | protein binding | | BTB/POZ fold | |
| 2530330 | 2.472 | | | | |
| 2606067 | 2.443 | | | | |
| 2600797 | 2.422 | ER retention sequence binding | protein-ER retention | ER lumen protein retaining receptor | |
| 2744292 | 2.345 | catalytic activity, transporter activity, triacylglycerol lipase activity | lipid metabolism, transport | Lipase, active site; Major intrinsic protein | |
| 2680842 | 2.065 | steroid metabolism | | Oxysterol-binding protein | |
| 2678866 | 6.701 | | | | |
| 2600178 | 6.547 | beta-catenin binding | development | Beta-catenin-interacting ICAT | |
| 2628500 | 5.752 | | | | |
| 2598981 | 5.595 | zinc ion binding | | Zinc finger, MYND-type; Batten's disease protein Cln3 | |
| 2681771 | 4.063 | NADPH binding aspartate-semialdehyde dehydrogenase activity | amino acid metabolism | Aspartate-semialdehyde dehydrogenase, conserved site | |
| 2602443 | 3.733 | nucleic acid binding | | RNA recognition motif, RNP-1 | |
| 2020262 | 3.627 | transferase activity, binding, acyltransferase activity | | Trimeric LpxA-like; Armadillo-type fold; Bacterial transferase hexapeptide repeat | |
| 2603816 | 3.543 | protein binding, binding | RNA metabolism | MIF4G-like, type 2; Armadillo-type fold | |
| 2747036 | 3.504 | zinc ion binding | | Zinc finger, MYND-type | |
| 1137889 | 3.169 | RNA processing, mRNA processing | | RNA-processing protein, HAT helix; Suppressor of forked | |
| 2674206 | 2.961 | nucleic acid binding, iron ion binding, ATP binding, helicase activity, catalase activity, heme binding, RNA binding, ribonuclease III activity, ATP dependent helicase activity | electron transport, RNA processing, response to oxidative stress | DNA/RNA helicase, DEAD/DEAH box type; Catalase; Ribonuclease III | |
| 2681774 | 2.921 | helicase activity, aminopeptidase activity, nucleic acid binding, ATP dependent helicase activity, ATP binding | | DNA/RNA helicase; Peptidase M17, leucyl aminopeptidase | |
| 2679584 | 2.333 | ubiquitin-protein ligase activity, nucleic acid binding, nuclease activity | protein modification, ubiquitin cycle | Parkin; Staphylococcal nuclease (SNase-like); Tudor; Ubiquitin | |
| 2680463 | 2.229 | helicase activity, catalytic activity, CTP synthase activity, nucleic acid binding, ATP dependent helicase activity; ATP binding | pyrimidine nucleotide biosynthesis | DNA/RNA helicase;Glutamine amidotransferase class-I; CTP synthase | |
| 2485812 | 2.184 | structural constituent of ribosome | protein biosynthesis | Ribosomal protein S12e | |
| 2702302 | 2.150 | nucleic acid binding | | Polynucleotidyl transferase, Ribonuclease H fold | |

| | | | | | |
|---|---|---|---|---|---|
| 2675285 | 2.139 | ATP binding, nucleotide binding, tRNA ligase activity, cysteine-tRNA ligase activity | cysteinyl-tRNA aminoacylation, amino acid activation, protein biosynthesis, transcription | | cytoplasm |
| 2750840 | 7.958 | | | | |
| 1188538 | 7.861 | arginase activity, hydrolase activity, acting on carbon-nitrogen (but not peptide) bonds, in linear amidines, metal ion binding | arginine catabolism | Arginase; Ureohydrolase | Metabolism |
| 2199166 | 7.660 | amino acid transporter activity | amino acid transport, transport | Amino acid/polyamine transporter I | |
| 2678898 | 7.006 | guanyl nucleotide binding, carbon-nitrogen ligase activity, with glutamine as amido-N-donor, signal transducer activity | signal transduction, G-protein coupled receptor protein signaling pathway | Guanine nucleotide binding protein (G-protein), alpha subunit; Amidase signature enzyme | |
| 2625518 | 6.886 | carboxy-lyase activity, pyridoxal phosphate binding | carboxylic acid metabolism | Pyridoxal phosphate-dependent decarboxylase | |
| 2680505 | 6.772 | monooxygenase activity, heme binding, iron ion binding | electron transport | Cytochrome P450 | |
| 2466833 | 6.693 | catalytic activity, FAD binding, oxidoreductase activity | | FAD-binding, type 2 | |
| 2667549 | 6.392 | monooxygenase activity, heme binding, iron ion binding | electron transport | Cytochrome P450 | |
| 2675740 | 6.295 | | | | |
| 2258133 | 5.500 | | metabolism | Aminotransferase, class V/Cysteine desulphurase | |
| 2597924 | 5.417 | catalytic activity, binding, oxidoreductase activity | metabolism | Flavoprotein; Short-chain dehydrogenase/reductase SDR; Glucose/ribitol dehydrogenase | |
| 2472473 | 5.078 | transferase activity, transferring alkyl or aryl (other than methyl) groups, pyridoxal phosphate binding | amino acid metabolism | Cys/Met metabolism, pyridoxal phosphate-dependent enzyme; O-acetylhomoserine/O-acetylserine sulfhydrylase | |
| 2443639 | 4.888 | lactoylglutathione lyase activity | carbohydrate metabolism | Glyoxalase I | |
| 2679758 | 4.887 | hydrolase activity, hydrolyzing O-glycosyl compounds | carbohydrate metabolism | Glycoside hydrolase family 2, immunoglobulin-like beta-sandwich | |
| 1118808 | 4.714 | catalytic activity | | Phosphatidic acid phosphatase type 2/haloperoxidase | |
| 2679876 | 4.430 | catalytic activity, cofactor binding, transporter activity, binding | transport, metabolism | AMP-dependent synthetase and ligase; Phosphopantetheine-binding; Major facilitator superfamily | |
| 2498267 | 4.403 | copper ion binding, amine oxidase activity, quinone binding | amine metabolism | Copper amine oxidase | |
| 2674336 | 4.244 | glycolipid 2-alpha-mannosyltransferase activity | protein amino acid glycosylation | Glycosyl transferase, family 15 | |
| 2600367 | 4.155 | FAD binding, iron ion binding, oxidoreductase activity | electron transport | Ferric reductase-like transmembrane component | |

| | | | | | |
|---|---|---|---|---|---|
| 2596642 | 3.941 | nucleoside triphosphatase activity, nucleotide binding, ATPase activity coupled to transmembrane movement of substances, ATPase activity, ATP binding | transport | AAA+ ATPase, core; ABC transporter, transmembrane region | |
| 2603600 | 3.939 | catalytic activity | cell wall catabolism, metabolism | Dihydroxy-acid and 6-phosphogluconate dehydratase; Peptidoglycan-binding LysM | |
| 2643580 | 3.876 | transporter activity, citrate transporter activity | sodium ion transport, citrate transport | Sodium/sulphate symporter; Divalent ion symporter | |
| 2601300 | 3.818 | catalytic activity, zinc ion binding, oxidoreductase activity | metabolism | Alcohol dehydrogenase, zinc-containing, conserved site | |
| 2552181 | 3.747 | oxidoreductase activity | electron transport | Taurine catabolism dioxygenase TauD/TfdA | |
| 2550951 | 3.557 | catalytic activity, binding | metabolism | NAD(P)-binding | |
| 2679197 | 3.541 | hydrolase activity, histone deacetylase activity | histone deacetylation | Metallophosphoesterase; Histone deacetylase | |
| 2681580 | 3.526 | nucleoside triphosphatase activity, nucleotide binding, ATPase activity, ATP binding | | AAA+ ATPase, core; ABC transporter, transmembrane region | |
| 2436028 | 3.479 | chorismate synthase activity | aromatic amino acid family biosynthesis | Chorismate synthase | |
| 2744134 | 3.477 | transferase activity, transferring hexosyl groups | carbohydrate biosynthesis | Glycosyl transferase, family 8 | |
| 2701913 | 3.329 | | | | |
| 2681970 | 3.317 | catalytic activity | metabolism | AMP-dependent synthetase and ligase | |
| 2091788 | 3.260 | NADH dehydrogenase ubiquinone) activity | electron transport | NADH-ubiquinone oxidoreductase B18 subunit | |
| 2596075 | 3.251 | nucleic acid binding, pyridoxal phosphate binding | cysteine metabolism, metabolism | RNA recognition motif, RNP-1; Cysteine desulphurase | |
| 2676524 | 3.222 | deaminase activity, AMP deaminase activity | purine ribonucleoside monophosphate biosynthesis | Adenosine/AMP deaminase active site | |
| 2595653 | 3.177 | kynureninase activity, pyridoxal phosphate binding, oxidoreductase activity | tryptophan catabolism, nicotinamide adenine dinucleotide biosynthesis, metabolism | Kynureninase; Aromatic-ring hydroxylase; FAD dependent oxidoreductase | |
| 2667262 | 3.164 | hydrolase activity, acting on carbon-nitrogen (but not peptide) bonds, in cyclic amides | pyrimidine base biosynthesis | Dihydroorotase, conserved site | |
| 2621172 | 3.074 | | | | |
| 2700718 | 3.049 | transaminase activity, pyridoxal phosphate binding | arginine metabolism | Aminotransferase class-III; Acetylornithine and succinylornithine aminotransferase | |
| 2440737 | 3.043 | hydrolase activity | | Amidohydrolase 1 | |
| 2525861 | 2.886 | imidazoleglycerol-phosphate dehydratase activity | histidine biosynthesis | Imidazole glycerol-phosphate dehydratase | |
| 2531122 | 2.843 | metallopeptidase activity, binding, zinc ion binding | proteolysis and peptidolysis, leukotriene biosynthesis | Armadillo-type fold; Peptidase M1 | |

| | | | | | |
|---|---|---|---|---|---|
| 2690581 | 2.565 | catalytic activity, calcium ion binding, alpha-amylase activity | carbohydrate metabolism | Glycoside hydrolase family 13; Alpha-amylase, fungi | |
| 2675588 | 2.399 | hydrolase activity, hydrolyzing O-glycosyl compounds, dihydrofolate reductase activity | nucleotide biosynthesis, lycine biosynthesis, carbohydrate metabolism | Glycoside hydrolase, clan GH-D; Dihydrofolate reductase region | |
| 2679939 | 2.377 | chitinase activity, hydrolase activity, hydrolyzing O-glycosyl compounds | chitin catabolism, carbohydrate metabolism | Chitinase II; Glycoside hydrolase, family 18, catalytic domain | |
| 2510648 | 2.334 | catalytic activity, hydrolase activity, zinc ion binding | | CMP/dCMP deaminase, zinc-binding | |
| 2600607 | 2.301 | | | | |
| 2494667 | 2.134 | | | | membrane |
| 2677880 | 2.086 | nucleoside triphosphatase activity, nucleotide binding, hedgehog receptor activity, ATP binding | | | integral to membrane, membrane |
| 2749150 | 10.230 | | | Allergen V5/Tpx-1 related | |
| 2308933 | 9.378 | | | Allergen V5/Tpx-1 related | |
| 2695463 | 7.525 | | | | |
| 2611041 | 6.945 | | | | Poorly characterized |
| 2660823 | 6.549 | metalloendopeptidase activity | proteolysis and peptidolysis | Peptidase M35, deuterolysin | |
| 2679537 | 6.372 | hydrolase activity, hydrolyzing O-glycosyl compounds, RNA binding, nucleic acid binding | RNA processing, carbohydrate metabolism | Glycoside hydrolase, family 43; SWAP/Surp; RNA recognition motif, RNP-1 | |
| 2598115 | 5.125 | transporter activity | transport | Major facilitator superfamily | |
| 2674161 | 4.763 | transition metal ion binding, hydrolase activity, oxidoreductase activity | | Ferritin/ribonucleotide reductase-like; Beta-lactamase-like | |
| 2110112 | 4.548 | | | Auxin efflux carrier | |
| 2611195 | 4.509 | | | | |
| 2717187 | 4.133 | | | | |
| 2587232 | 3.959 | sugar porter activity, transporter activity | carbohydrate transport, transport | General substrate transporter; Major facilitator superfamily; Sugar transporter, conserved site | |
| 2748760 | 3.942 | | | | |
| 2680216 | 3.891 | nucleic acid binding, zinc ion binding | | Zinc finger, C2H2-type | |
| 2195169 | 3.784 | | | | |
| 2289565 | 3.670 | metalloendopeptidase activity, metallopeptidase activity, zinc ion binding | proteolysis and peptidolysis | Peptidase M48, Ste24p | |
| 2584235 | 3.663 | phosphoric monoester hydrolase activity | metabolism | HAD-superfamily hydrolase, subfamily IB, PSPase-like | |
| 2321290 | 3.487 | DNA binding, transcription factor activity, zinc ion binding | transcription, regulation of transcription, DNA-dependent | Fungal specific transcription factor | |
| 76079 | 3.479 | sugar porter activity, transporter activity | carbohydrate transport, transport | Sugar transporter, conserved site; General substrate transporter; Major facilitator superfamily | |
| 2274818 | 3.062 | nucleic acid binding | | RNA recognition motif, RNP-1 | |

| | | | | | |
|---|---|---|---|---|---|
| 2675502 | 3.039 | | | | |
| 2596945 | 3.022 | hydrogen-transporting ATPase activity, rotational mechanism, protein binding, hydrogen-transporting ATP synthase activity, rotational mechanism, zinc ion binding | ATP synthesis coupled proton transport | ATPase, F1 complex, OSCP/delta subunit; Zinc finger, RING-type | |
| 2680910 | 2.881 | RNA binding, nucleic acid binding | RNA processing | Lupus La protein; RNA recognition motif, RNP-1 | |
| 2602357 | 2.360 | protein binding | amino acid transport, transport | BTB/POZ fold; Amino acid permease, conserved site | |
| 2599506 | 2.317 | C-8 sterol isomerase activity | ergosterol biosynthesis | ERG2 and sigma1 receptor-like | |
| 2681478 | 2.310 | nucleic acid binding, zinc ion binding | | Zinc finger, CCCH-type; RNA recognition motif, RNP-1 | |
| 2680499 | 2.292 | cytochrome-c oxidase activity | electron transport | Cytochrome c oxidase, subunit Vib | |
| 2579584 | 10.438 | subtilase activity, serine-type peptidase activity | proteolysis and peptidolysis | Peptidase S8 and S53, subtilisin, kexin, sedolisin | |
| 2481522 | 10.429 | | | | |
| 2598518 | 10.117 | | | | |
| 2467818 | 9.867 | | | | |
| 2635172 | 9.764 | | | | |
| 2549292 | 9.610 | | | | |
| 2618740 | 9.580 | | | | |
| 2541149 | 9.544 | | | | |
| 2470835 | 9.333 | | | | |
| 2678339 | 9.200 | | | | |
| 2541426 | 9.161 | | | | |
| 2295360 | 9.084 | | | | |
| 2626480 | 9.056 | | | | |
| 2493980 | 8.846 | | | | |
| 2602724 | 8.714 | | | | |
| 2701497 | 8.629 | | | | |
| 2703131 | 8.331 | | | | |
| 2534073 | 8.274 | | | | |
| 2696974 | 7.884 | RNA-directed DNA polymerase activity, nucleic acid binding, zinc ion binding, DNA binding, aspartic-type endopeptidase activity, RNA binding | RNA dependent DNA replication, DNA integration, proteolysis and peptidolysis | RNA-directed DNA polymerase (reverse transcriptase); Zinc finger, CCHC-type; Polynucleotidyl transferase, Ribonuclease H fold; Zinc finger, CCHC-type; Polynucleotidyl transferase; Integrase, catalytic core | |
| 2663658 | 7.804 | | | | |
| 2745841 | 7.679 | | | | |
| 2442813 | 7.645 | | | | |
| 2750895 | 7.613 | | | | |
| 2586726 | 7.557 | | | | |
| 2541924 | 7.393 | | | | |
| 2599691 | 7.346 | | | | |
| 2350680 | 7.342 | | | | |
| 2197213 | 7.327 | | | | |
| 2597881 | 7.284 | aspartic-type endopeptidase activity, pepsin A activity | lipid metabolism, metabolism, proteolysis and peptidolysis | Peptidase aspartic, catalytic; Patatin; Acyl transferase/acyl hydrolase/lysophospholipase | |

| | | | | | |
|---|---|---|---|---|---|
| 2636155 | 7.199 | ribonuclease activity, endoribonuclease activity, RNA binding | | Ribonuclease/ribotoxin; Guanine-specific ribonuclease N1 and T1 | |
| 2749147 | 7.071 | | | Allergen V5/Tpx-1 related | |
| 2570153 | 6.838 | | | | |
| 2638754 | 6.707 | FAD binding, pyranose oxidase activity, oxidoreductase activity, acting on CH-OH group of donors, oxidoreductase activity | glucose metabolism | Glucose-methanol-choline oxidoreductase; Pyranose oxidase; FAD dependent oxidoreductase | |
| 2584950 | 6.412 | | | | |
| 2519451 | 6.325 | | | | |
| 2602828 | 6.109 | catalytic activity, hydrolase activity, acting on carbon-nitrogen (but not peptide) bonds | carbohydrate metabolism | Polysaccharide deacetylase; Glycoside hydrolase/deacetylase, beta/alpha-barrel | |
| 2667748 | 6.010 | | | | |
| 2317702 | 5.896 | nucleic acid binding, zinc ion binding | | Zinc finger, C2H2-type | |
| 1187227 | 5.879 | | | | |
| 2677004 | 5.876 | | | | |
| 2503584 | 5.559 | | | | |
| 2157839 | 5.239 | | | | |
| 2685946 | 5.185 | oxidoreductase activity | electron transport | Amine oxidase | |
| 2545442 | 5.055 | hydrolase activity, hydrolyzing O-glycosyl compounds | carbohydrate metabolism | Glycoside hydrolase, family 3 | |
| 2639888 | 4.922 | | | | |
| 2506333 | 4.906 | | | | |
| 2556613 | 4.859 | | | | |
| 2017878 | 4.726 | | | | |
| 2681337 | 4.616 | actin binding | cytoskeleton organization and biogenesis | Profilin/allergen | |
| 2680606 | 4.495 | | | | |
| 2581457 | 4.383 | | | | |
| 2675539 | 4.256 | transcription factor activity, nucleic acid binding, zinc ion binding | regulation of transcription, DNA-dependent | Fungal transcriptional regulatory protein; Zinc finger, CCHC-type | |
| 2625719 | 4.227 | aminopeptidase activity, prolyl aminopeptidase activity | proteolysis and peptidolysis | Peptidase S33, prolyl aminopeptidase | |
| 2601114 | 4.193 | transcription factor activity, zinc ion binding | regulation of transcription, DNA-dependent | Fungal transcriptional regulatory protein, N-terminal | |
| 2701101 | 4.144 | oxidoreductase activity | metabolism | Aldehyde/histidinol dehydrogenase | |
| 2681665 | 4.111 | | | | |
| 2601373 | 4.044 | | | | |
| 2677655 | 3.971 | DNA binding | | Myb, DNA-binding | |
| 2598849 | 3.921 | hydrolase activity | | Ham1-like protein | |
| 1097623 | 3.915 | | | | |
| 2595539 | 3.873 | | | | |
| 2602716 | 3.765 | peroxidase activity, heme binding | electron transport, response to oxidative stress | Haem peroxidase | |
| 2516775 | 3.725 | | | | |

| | | | | | |
|---|---|---|---|---|---|
| 2597288 | 3.583 | FAD binding, oxidoreductase activity, acting on CH-OH group of donors | | Glucose-methanol-choline oxidoreductase | |
| 2546907 | 3.544 | carbohydrate binding, catalytic activity, glucan 1,4-alpha-glucosidase activity | polysaccharide catabolism, polysaccharide metabolism, carbohydrate metabolism | Carbohydrate-binding-like fold; Glycoside hydrolase, carbohydrate-binding; Six-hairpin glycosidase-like | |
| 2743569 | 3.515 | | | | |
| 2603514 | 3.289 | nucleic acid binding | | RNA recognition motif, RNP-1 | |
| 2367013 | 3.248 | | | | |
| 2688935 | 3.223 | | | | |
| 2707024 | 3.171 | structural constituent of ribosome | protein biosynthesis | Ribosomal protein L19/L19e | |
| 2680244 | 3.148 | nucleic acid binding | | RNA recognition motif, RNP-1 | |
| 2700082 | 3.131 | zinc ion binding | | Zinc finger, MYND-type | Poorly characterized |
| 2538638 | 3.093 | catalytic activity, binding, oxidoreductase activity | metabolism | NAD(P)-binding; Short-chain dehydrogenase/reductase SDR; Glucose/ribitol dehydrogenase | |
| 2018047 | 3.081 | peroxidase activity | electron transport | Chloroperoxidase | |
| 2449841 | 3.046 | protein binding | | BTB/POZ fold | |
| 1199149 | 3.013 | | | | |
| 2632337 | 3.009 | | | | |
| 2680023 | 2.908 | hydrolase activity, acting on carbon-nitrogen (but not peptide) bonds, chorismate synthase activity, hydrolase activity | aromatic amino acid family biosynthesis | Metal-dependent hydrolase, composite; Chorismate synthase; Amidohydrolase 1 | |
| 2746514 | 2.886 | ATP binding, zinc ion binding, DNA binding, NAD synthase (glutamine-hydrolyzing) activity, hydrolase activity, acting on carbon-nitrogen (but not peptide) bonds | transcription, nicotinamide adenine dinucleotide biosynthesis, nitrogen metabolism | Fungal specific transcription factor; NAD+ synthase; Nitrilase/cyanide hydratase and apolipoprotein N-acyltransferase | |
| 2677660 | 2.821 | zinc ion binding | | Zinc finger, B-box | |
| 2517501 | 2.804 | oxidoreductase activity, acting on single donors with incorporation of molecular oxygen, incorporation of two atoms of oxygen, catalytic activity, ferric iron binding, ron ion binding | aromatic compound metabolism | Intradiol ring-cleavage dioxygenase, core | |
| 2468516 | 2.793 | | | | |
| 2676403 | 2.725 | chorismate mutase activity | aromatic amino acid family biosynthesis | Chorismate mutase; Prohibitin | |
| 2674711 | 2.719 | hydrolase activity | | Beta-lactamase-like | |
| 2705231 | 2.676 | hydrolase activity | | NUDIX hydrolase, core | |
| 2679472 | 2.664 | protein binding, GTP binding, GTPase activity | nucleocytoplasmic transport, signal transduction, small GTPase mediated signal transduction, protein | Ras small GTPase, Ras type; Ras small GTPase, Rho type; Ras small GTPase, Rab type; Ran GTPase; Small GTP-binding protein | |

| | | | | | |
|---|---|---|---|---|---|
| | | | transport, intracellular protein transport | | |
| 2600031 | 2.631 | hydrolase activity, hydrolyzing O-glycosyl compounds | carbohydrate metabolism | Glycoside hydrolase, family 5 | |
| 2216136 | 2.557 | structural constituent of ribosome | translational elongation | Ribosomal protein 60S | |
| 2535493 | 2.548 | hydrolase activity, hydrolyzing O-glycosyl compounds | carbohydrate metabolism | Glycoside hydrolase, family 5 | |
| 2680434 | 2.547 | hydrolase activity, hydrolyzing O-glycosyl compounds | carbohydrate metabolism | Glycoside hydrolase, family 5 | |
| 2504553 | 2.447 | structural constituent of ribosome | protein biosynthesis | Ribosomal protein S2 | |
| 2677775 | 2.415 | DNA binding, monooxygenase activity, DNA-directed RNA polymerase activity, heme binding, iron ion binding | electron transport, transcription | Cytochrome P450; RNA polymerase | |
| 2677329 | 2.414 | protein binding, structural molecule activity, protein transporter activity | protein complex assembly, vesicle-mediated transport, intracellular protein transport | Coatomer, WD associated region | |
| 2458666 | 2.347 | calcium ion binding | protein folding | Calreticulin/calnexin | |
| 2152826 | 2.222 | ubiquinol-cytochrome-c reductase activity, oxidoreductase activity | electron transport | Ubiquinol cytochrome reductase transmembrane region; Rieske iron-sulphur protein | |
| 2527041 | 2.195 | nucleic acid binding, zinc ion binding | | Zinc finger, CCHC-type | |
| 2587192 | 2.156 | actin binding | | Actinin-type, actin-binding, conserved site | Cellular processes and signaling |
| 2675127 | 2.039 | ubiquitin-like-protein ligase activity | | Ubiquitin-conjugating enzyme, E2 | |

## Table S21. Repressed proteins in fruiting bodies vs. monokaryon 12-43

| Schco3 ID | Fold changes | Go Desc molecular function | Go Desc Biological Process | Interpro Desc | KOG Group |
|---|---|---|---|---|---|
| 2746877 | -2.025 | protein binding, structural constituent of ribosome, zinc ion binding | protein biosynthesis | Ribosomal protein L5 | |
| 2725707 | -2.040 | protein kinase activity, protein-tyrosine kinase activity, protein serine/threonine kinase activity, ATP binding | protein amino acid phosphorylation | Protein kinase; Serine/threonine protein kinase-related; Tyrosine protein kinase | |
| 2675684 | -2.098 | metalloendopeptidase activity, ATP binding, zinc ion binding, nucleotide binding, nucleoside triphosphatase activity | protein catabolism, proteolysis and peptidolysis | AAA ATPase; Peptidase M41 | |
| 2589186 | -2.102 | protein transporter activity, binding | protein-nucleus import, docking, intracellular protein transport | Importin-beta; Armadillo-type fold | |
| 2478805 | -2.103 | signal transducer activity | intracellular signaling cascade | Regulator of G protein signalling; Pleckstrin/ G-protein, interacting region | |

| | | | | | |
|---|---|---|---|---|---|
| 2504769 | -2.107 | catalytic activity | | Prenyltransferase/squalene oxidase | |
| 2555639 | -2.121 | | | | |
| 2601822 | -2.127 | catalytic activity, zinc ion binding | | Six-hairpin glycosidase-like | |
| 2491587 | -2.132 | ubiquitin-like-protein ligase activity | | Ubiquitin-conjugating enzyme, E2 | |
| 2694313 | -2.176 | | | | |
| 2597001 | -2.201 | | | | |
| 2600099 | -2.207 | oxidoreductase activity | | Redoxin | |
| 2600947 | -2.233 | protein kinase activity, protein serine/threonine kinase activity, nucleic acid binding, ATP binding, protein-tyrosine kinase activity | protein amino acid phosphorylation , intracellular protein transport | Protein kinase; Serine/threonine protein kinase-related; Protein kinase ATP binding, conserved site | |
| 2266989 | -2.251 | | | | |
| 2674818 | -2.252 | catalytic activity, formate-tetrahydrofolate ligase activity, binding, 1,3-beta-glucan synthase activity, ATP binding | beta-1,3 glucan biosynthesis, folic acid and derivative biosynthesis, metabolism | Tetrahydrofolate dehydrogenase/cyclohydrolase | |
| 2528872 | -2.260 | protein kinase activity, protein-tyrosine kinase activity, protein serine/threonine kinase activity, ATP binding | protein amino acid phosphorylation | Protein kinase; Serine/threonine protein kinase-related; Protein kinase ATP binding, conserved site | |
| 2207599 | -2.294 | actin binding | | Actin-binding, cofilin/tropomyosin type | |
| 2682064 | -2.299 | protein binding | protein complex assembly, transport, vesicle-mediated transport, intracellular protein transport | Clathrin adaptor, sigma subunit/coatomer, zeta subunit; Longin-like | |
| 2701639 | -2.349 | threonine endopeptidase activity | ubiquitin-dependent protein catabolism | 20S proteasome, A and B subunits | |
| 2594767 | -2.354 | protein binding, ATP binding | | Actin/actin-like | |
| 2599395 | -2.414 | catalytic activity, small protein activating enzyme activity | protein modification, ubiquitin cycle | Ubiquitin-activating enzyme, E1 | |
| 2482990 | -2.463 | | | | |
| 2056522 | -2.468 | DNA binding | | Histone-fold | |
| 2544442 | -2.558 | protein kinase activity, protein-tyrosine kinase activity, protein serine/threonine kinase activity, ATP binding | protein amino acid phosphorylation | Protein kinase; Serine/threonine protein kinase-related; Protein kinase ATP binding, conserved site | |
| 2709241 | -2.561 | ATP binding | protein folding | ATP-binding region, ATPase-like; Heat shock protein Hsp90 | |
| 2704716 | -2.569 | nucleoside triphosphatase activity, nucleotide binding, hydrolase activity, ATP binding | protein catabolism | AAA+ ATPase, core; 26S proteasome subunit P45 | |
| 2753824 | -2.635 | steroid metabolism | | Oxysterol-binding protein | |
| 2597433 | -2.651 | protein kinase activity, protein-tyrosine kinase activity, protein | protein amino acid phosphorylation | Protein kinase; Serine/threonine protein kinase-related; Tyrosine protein kinase | |

| | | | | | |
|---|---|---|---|---|---|
| | | serine/threonine kinase activity, ATP binding | | | |
| 2677294 | -2.661 | catalytic activity, binding, coenzyme binding | metabolism | NAD-dependent epimerase/dehydratase | |
| 2598895 | -2.669 | protein binding | cell communication | Phox-like | |
| 2531252 | -2.707 | UDP-glucose 4-epimerase activity, catalytic activity, binding, coenzyme binding | metabolism, galactose metabolism | UDP-glucose 4-epimerase | |
| 2016319 | -2.725 | protein binding, GTP binding, GTPase activity | nucleocytoplasmic transport, signal transduction, small GTPase mediated signal transduction, protein transport, intracellular protein transport | Ras small GTPase, Ras type; Ras small GTPase, Rho type; Ras small GTPase, Rab type; Small GTP-binding protein | |
| 2481091 | -2.749 | | | | |
| 2689527 | -2.798 | 1,3-beta-glucan synthase activity | beta-1,3 glucan biosynthesis | Glycosyl transferase, family 48 | |
| 1157296 | -2.807 | ATP binding | | Heat shock protein Hsp70 | |
| 2535557 | -2.812 | regulation of actin polymerization | | Arp2/3 complex, 34kDa subunit p34-Arc | |
| 2453944 | -2.816 | phosphoric monoester hydrolase activity, protein tyrosine/serine/threonine phosphatase activity, protein tyrosine phosphatase activity | protein amino acid dephosphorylation, dephosphorylation | Protein-tyrosine phosphatase | |
| 2572877 | -2.862 | peptidase activity | proteolysis and peptidolysis | Peptidase M28 | |
| 2666833 | -2.894 | nucleoside metabolism | | Phosphoribosyltransferase | |
| 2498761 | -2.924 | | | | |
| 2354629 | -2.941 | GTP binding | protein transport, small GTPase mediated signal transduction, intracellular protein transport | ARF/SAR superfamily; Ras GTPase; GTP-binding protein SAR1; Small GTP-binding protein | Cellular processes and signaling |
| 2679029 | -2.984 | protein kinase CK2, intrinsic regulator activity, DNA binding | protein folding | Casein kinase II, regulatory subunit; Histone-fold; Heat shock protein DnaJ; Transcription factor CBF/NF-Y/archaeal histone | |
| 2110939 | -2.996 | protein folding | | Heat shock protein DnaJ | |
| 2674535 | -3.006 | protein binding, ATP binding | protein folding | Chaperonin Cpn60; T-complex protein 1; GroEL-like chaperone, ATPase | |
| 2697890 | -3.029 | protein kinase activity, protein-tyrosine kinase activity, protein serine/threonine kinase activity, ATP binding | intracellular signaling cascade, signal transduction, protein amino acid phosphorylation | Protein kinase; Serine/threonine protein kinase-related; Protein kinase ATP binding, conserved site | |
| 2574819 | -3.040 | | | | |
| 2458589 | -3.043 | | | | |
| 2599910 | -3.047 | protein binding | | Actin/actin-like | |
| 2602808 | -3.116 | protein prenyltransferase activity | protein amino acid prenylation | Protein prenyltransferase, alpha subunit | |

| | | | | | |
|---|---|---|---|---|---|
| 2676846 | -3.116 | protein binding, structural molecule activity, binding, protein transporter activity | protein complex assembly, intracellular protein transport, vesicle-mediated transport | Clathrin, heavy chain/VPS, 7-fold repeat | |
| 2706648 | -3.142 | protein binding, protein transporter activity | protein complex assembly, transport, vesicle-mediated transport, intracellular protein transport | Clathrin adaptor, mu subunit | |
| 2596663 | -3.154 | nucleotidyltransferase activity | metabolism | UTP--glucose-1-phosphate uridylyltransferase | |
| 2674968 | -3.165 | nucleic acid binding, ATP binding, zinc ion binding | | Zinc finger, C2H2-type | |
| 2676856 | -3.209 | calcium ion binding, DNA binding, nucleotide-sugar transporter activity, oxidoreductase activity | electron transport, nucleotide-sugar transport | Calcium-binding EF-hand; SANT, DNA-binding; UDP-galactose transporter; Flavin-containing amine oxidase | |
| 2685615 | -3.223 | actin binding | actin cytoskeleton organization and biogenesis | F-actin capping protein, alpha subunit | |
| 2188670 | -3.229 | catalytic activity, metalloendopeptidase activity, metal ion binding, zinc ion binding | proteolysis and peptidolysis | Metalloenzyme, LuxS/M16 peptidase-like, metal-binding | |
| 2249278 | -3.233 | | | | |
| 2488292 | -3.248 | | | | |
| 2599106 | -3.284 | nucleoside triphosphatase activity, nucleotide binding, serine-type peptidase activity, binding, ATP binding | | AAA+ ATPase, core; Peptidase S53, propeptide; Aspartate decarboxylase-like fold; 1 Cell division protein 48 | |
| 2616372 | -3.338 | protein transporter activity, binding | protein-nucleus import, docking, intracellular protein transport | Importin-beta; Armadillo-type fold | |
| 2600008 | -3.348 | protein binding, structural molecule activity, binding, transporter activity, protein transporter activity | protein complex assembly, transport, intracellular protein transport, vesicle-mediated transport | Clathrin/coatomer adaptor, adaptin-like; Coatomer, beta subunit, Armadillo-type fold; Major facilitator superfamily | |
| 2175961 | -3.387 | protein kinase activity, MAP kinase activity, protein-tyrosine kinase activity, protein serine/threonine kinase activity, ATP binding | protein amino acid phosphorylation | Protein kinase ATP binding, conserved site; MAP kinase, conserved site; Tyrosine protein kinase; Serine/threonine protein kinase | |
| 2596581 | -3.392 | protein binding | cell communication | Phox-like | |
| 2492200 | -3.406 | serine-type peptidase activity | proteolysis and peptidolysis | Peptidase S28 | |
| 2709667 | -3.427 | protein binding, motor activity, ATP binding | | Myosin S1 fragment; Myosin head | |
| 2599999 | -3.442 | catalytic activity, binding, protein transporter | cell proliferation, protein-nucleus | Phosphatidic acid phosphatase type 2/haloperoxidase; Armadillo-type fold; Importin-beta; CAS/CSE | |

| | | | | | |
|---|---|---|---|---|---|
| | | activity, importin-alpha export receptor activity | import, docking, apoptosis, intracellular protein transport | | |
| 2601777 | -3.444 | autophagy | | Autophagy protein 5 | |
| 2349132 | -3.467 | protein kinase activity, protein-tyrosine kinase activity, protein serine/threonine kinase activity, ATP binding | protein amino acid phosphorylation | Protein kinase; Serine/threonine protein kinase-related; Protein kinase ATP binding, conserved site | |
| 2675504 | -3.553 | metalloexopeptidase activity, hydrolase activity, methionyl aminopeptidase activity | metabolism, proteolysis and peptidolysis | Alpha/beta hydrolase fold-3; Lipase, GDXG, active site; Peptidase M24 | |
| 2603003 | -3.643 | ubiquitin-like-protein ligase activity | | Ubiquitin-conjugating enzyme, E2 | |
| 2600989 | -3.664 | ATP binding, binding, DNA binding, DNA topoisomerase type II activity | signal transduction, DNA metabolism, transport, DNA topological change | Spo11/DNA topoisomerase VI, subunit A; Mitochondrial substrate carrier; Rho GAP | |
| 2482758 | -3.684 | nucleoside triphosphatase activity, nucleotide binding, hydrolase activity, ATP binding | protein catabolism | AAA ATPase; 26S proteasome subunit P45 | |
| 2127007 | -3.773 | metal ion binding | | Class II aldolase/adducin | |
| 2458250 | -3.796 | peptidase activity | proteolysis and peptidolysis | Peptidase M28 | |
| 2161007 | -3.873 | protein binding, binding, protein transporter activity | protein complex assembly, intracellular protein transport, vesicle-mediated transport | Clathrin/coatomer adaptor, adaptin-like; Armadillo-type fold | |
| 2601514 | -3.960 | protein binding, ATP binding | protein folding | Chaperonin Cpn60/TCP-1; T-complex protein 1, zeta subunit | |
| 2545132 | -3.963 | GTPase activity, structural molecule activity, GTP binding | microtubule-based movement, microtubule-based process | Tubulin/FtsZ, C-terminal; Beta tubulin | |
| 2601975 | -4.052 | | | | |
| 2576552 | -4.066 | protein kinase activity, protein-tyrosine kinase activity, protein serine/threonine kinase activity, ATP binding | protein amino acid phosphorylation | Protein kinase; Serine/threonine protein kinase-related; Tyrosine protein kinase | |
| 2304838 | -4.082 | protein folding | | Chaperone DnaJ; Heat shock protein DnaJ; HSP40/DnaJ peptide-binding | |
| 2685426 | -4.181 | protein binding, ATP binding | protein folding | Chaperonin Cpn60/TCP-1; T-complex protein 1, gamma subunit; Chaperonin TCP-1, conserved site; GroEL-like chaperone, ATPase | |
| 2744020 | -4.202 | protein binding, GTP binding, GTPase activity | nucleocytoplasmic transport, signal transduction, small GTPase mediated signal transduction, protein | Ras small GTPase, Ras type; Ras small GTPase, Rho type; Ras small GTPase, Rab type; Ran GTPase; Small GTP-binding protein | |

| | | | | | |
|---|---|---|---|---|---|
| | | | transport, intracellular protein transport | | |
| 2464773 | -4.229 | protein binding | | Actin/actin-like | |
| 2595604 | -4.273 | catalytic activity, Rab GTPase activator activity, binding, oxidoreductase activity | metabolism | RabGAP/TBC | |
| 2247557 | -4.273 | | | | |
| 1141646 | -4.280 | GTPase activity, structural molecule activity, GTP binding | microtubule-based movement, microtubule-based process | Tubulin/FtsZ; Alpha tubulin | |
| 2603876 | -4.283 | | | | |
| 2680097 | -4.302 | acid phosphatase activity, zinc ion binding, transcription factor activity, ARF guanyl-nucleotide exchange factor activity | regulation of transcription, DNA-dependent | Histidine acid phosphatase; SEC7-like | |
| 2596185 | -4.336 | protein binding, GTP binding, GTPase activity | nucleocytoplasmic transport, signal transduction, small GTPase mediated signal transduction, protein transport, intracellular protein transport | Ras small GTPase, Ras type; Ras small GTPase, Rho type; Ras small GTPase, Rab type; Ran GTPase; Small GTP-binding protein | |
| 2723169 | -4.341 | protein transporter activity, binding | protein-nucleus import, docking, intracellular protein transport | Importin-beta; Armadillo-type fold | |
| 2480386 | -4.372 | protein binding, ATP binding | protein folding | Chaperonin Cpn60 | |
| 2481214 | -4.389 | carboxypeptidase A activity, zinc ion binding | proteolysis and peptidolysis | Peptidase M14, carboxypeptidase A | |
| 2602353 | -4.433 | | | | |
| 2750934 | -4.820 | protein folding | | Peptidyl-prolyl cis-trans isomerase, FKBP-type | |
| 2599215 | -4.856 | GTPase activity, structural molecule activity, GTP binding | microtubule-based movement, microtubule-based process | Alpha tubulin | |
| 2521613 | -4.867 | nucleoside metabolism | | Phosphoribosyltransferase | |
| 2675738 | -5.016 | intracellular protein transport | | NSF attachment protein | |
| 2702602 | -5.141 | calcium ion binding | | Calcium-binding EF-hand | |
| 2674326 | -5.279 | microtubule motor activity, ATP binding | microtubule-based movement | Kinesin, motor region | |
| 2041665 | -5.361 | | | | |
| 2597700 | -5.417 | nucleoside triphosphatase activity, nucleotide binding, hydrolase activity, ATP binding | protein catabolism | AAA ATPase; 26S proteasome subunit P45 | |
| 2754081 | -5.739 | antioxidant activity, oxidoreductase activity | Alkyl hydroperoxide reductase/ Thiol specific | | |

| | | | | | |
|---|---|---|---|---|---|
| | | | antioxidant/ Mal allergen | | |
| 2582680 | -5.762 | binding | | Armadillo-type fold | |
| 2678570 | -6.440 | protein binding, binding, peptidyl-prolyl cis-trans isomerase activity | protein folding, RNA metabolism | Peptidyl-prolyl cis-trans isomerase, cyclophilin-type; MIF4G-like, type 3; Armadillo-type fold | |
| 2500485 | -6.609 | chitin synthase activity | chitin biosynthesis | Chitin synthase | |
| 2678401 | -7.221 | GTPase activity, structural molecule activity, GTP binding | microtubule-based movement, microtubule-based process | Beta tubulin | |
| 2675082 | -9.529 | DNA binding, DNA-directed RNA polymerase activity | transcription | DNA-directed RNA polymerase, M/15 kDa subunit | |
| 2675703 | -2.101 | catalytic activity, succinate-CoA ligase (ADP-forming) activity, phosphotransferase activity, alcohol group as acceptor, binding, ATP citrate synthase activity, transferaseactivity, transferring phosphorus-containing groups acyl-CoA binding | metabolism | Phosphatidylinositol 3- and 4-kinase | |
| 2705009 | -2.118 | leucine-tRNA ligase activity, ATP binding, nucleotide binding, tRNA ligase activity | leucyl-tRNA aminoacylation, amino acid activation, protein biosynthesis | Valyl/Leucyl/Isoleucyl-tRNA synthetase, class I, anticodon-binding; Aminoacyl-tRNA synthetase, class I (M); Leucyl-tRNA synthetase, class Ia, archaeal/eukaryotic cytosolic | |
| 2600306 | -2.203 | ATP binding, threonine-tRNA ligase activity, nucleotide binding, ligase activity, forming aminoacyl-tRNA and related compounds, tRNA ligase activity | amino acid activation, protein biosynthesis, threonyl-tRNA aminoacylation | Threonyl-tRNA synthetase, class Iia; Aminoacyl-tRNA synthetase, class II (G, H, P and S); Anticodon-binding | |
| 2697041 | -2.214 | nucleic acid binding, ATP binding, binding, zinc ion binding, DNA binding, helicase activity, transporter activity, hydrolase activity | transport | DNA/RNA helicase; Restriction endonuclease, type I, R subunit/Type III; Mitochondrial carrier protein | |
| 2147637 | -2.251 | aspartate-tRNA ligase activity, nucleic acid binding, ATP binding, nucleotide binding, transporter activity, tRNA ligase activity | amino acid activation, protein biosynthesis, aspartyl-tRNA aminoacylation, transport | Aspartyl-tRNA synthetase, class Iib | |
| 2268949 | -2.282 | mRNA metabolism | | Like-Sm ribonucleoprotein | |
| 2680463 | -2.291 | helicase activity, catalytic activity, CTP synthase activity, nucleic acid , ATP dependent helicase activity, ATP binding | pyrimidine nucleotide biosynthesis | DNA/RNA helicase; Glutamine amidotransferase class-I; CTP synthase | |
| 2568544 | -2.472 | translation initiation factor activity | translational initiation | Translation initiation factor IF2/IF5 | |
| 2678650 | -2.516 | structural constituent of ribosome | protein biosynthesis | Ribosomal protein L31e | |
| 2596209 | -2.534 | helicase activity, nucleic acid binding, ATP dependent helicase activity, ATP binding | | DNA/RNA helicase | |

| | | | | | |
|---|---|---|---|---|---|
| 2548156 | -2.540 | alanine-tRNA ligase activity, ligase activity, forming aminoacyl-tRNA and related compounds, nucleic acid binding, ATP binding | alanyl-tRNA aminoacylation, protein biosynthesis | Alanyl-tRNA synthetase, class Iic; Threonyl/alanyl tRNA synthetase, SAD; Phosphoesterase, DHHA1 | |
| 2700057 | -2.549 | phenylalanine-tRNA ligase activity, ATP binding, nucleotide binding, transition metal ion binding, RNA binding, heme binding | phenylalanyl-tRNA aminoacylation, protein biosynthesis | B3/B4 tRNA-binding domain; Phenylalanyl-tRNA synthetase, class IIc, beta subunit, archae/euk cytosolic | |
| 2599769 | -2.558 | protein binding, GTP binding, nucleic acid binding, ATP binding, zinc ion binding, helicase activity, ATP dependent helicase activity | small GTPase mediated signal transduction, protein transport | Ras small GTPase, Ras type; Ras small GTPase, Rho type; Ras small GTPase, Rab type; Small GTP-binding protein | |
| 2620095 | -2.579 | | | | |
| 2681753 | -2.617 | ATP binding, nucleotide binding, histidine-tRNA ligase activity, tRNA ligase activity | histidyl-tRNA aminoacylation, amino acid activation, protein biosynthesis | Aminoacyl-tRNA synthetase, class II (G, H, P and S); Anticodon-binding; Histidyl-tRNA synthetase, class IIa, subgroup | |
| 2638742 | -2.649 | mRNA metabolism | | Like-Sm ribonucleoprotein | |
| 2595220 | -2.714 | farnesyl-diphosphate farnesyltransferase activity, transferase activity, zinc ion binding | biosynthesis, lipid biosynthesis | Farnesyl-diphosphate farnesyltransferase; Squalene/phytoene synthase | |
| 2523486 | -2.810 | tryptophan-tRNA ligase activity, ATP binding, nucleotide binding, tRNA ligase activity | tryptophanyl-tRNA aminoacylation, amino acid activation, protein biosynthesis | Tryptophanyl-tRNA synthetase, class Ib; Aminoacyl-tRNA synthetase, class Ib | |
| 2598148 | -2.856 | structural constituent of ribosome, ATP binding, DNA helicase activity | protein biosynthesis | Ribosomal protein S19/S15; AAA ATPase, core; TIP49 | |
| 2596995 | -2.904 | nucleic acid binding, ATP binding, DNA binding, helicase activity, hydrolase activity, hydrolyzing O-glycosyl compounds | carbohydrate metabolism | DNA/RNA helicase; SNF2-related | |
| 2166139 | -2.924 | structural constituent of ribosome | protein biosynthesis | Ribosomal protein S9 | |
| 2663340 | -2.997 | structural constituent of ribosome | protein biosynthesis | Ribosomal protein S16 | |
| 2743097 | -3.041 | catalytic activity, binding | carbohydrate metabolism | Glycosyl hydrolase, family 13, catalytic region; Armadillo-type fold | |
| 2598311 | -3.205 | argininosuccinate synthase activity, NAD-dependent histone deacetylase activity, ATP binding, zinc ion binding, DNA binding | arginine biosynthesis, regulation of transcription, DNA-dependent, protein amino acid deacetylation, chromatin silencing | Argininosuccinate synthase; NAD-dependent histone deacetylase, silent information regulator Sir2 | |
| 2528965 | -3.209 | double-stranded RNA binding, RNA binding, ribonuclease III activity | RNA processing | Double-stranded RNA binding; Ribonuclease III | |

| | | | | | |
|---|---|---|---|---|---|
| 2681298 | -3.311 | thiamin diphosphokinase activity, ATP binding, nucleotide binding, lycine-tRNA ligase activity, tRNA ligase activity | thiamin diphosphate biosynthesis, amino acid activation, thiamin metabolism, protein biosynthesis, glycyl-tRNA aminoacylation | Thiamin pyrophosphokinase, catalytic region; Glycyl-tRNA synthetase, alpha2 dimer; Aminoacyl-tRNA synthetase, class II (G, H, P and S); Anticodon-binding | |
| 2679031 | -3.313 | protein binding, nucleic acid binding, ATP binding, helicase activity, protein transporter activity, ATP dependent helicase activity | protein complex assembly, transport, intracellular protein transport, vesicle-mediated transport | Clathrin adaptor, mu subunit; DANN RNA Helicase | |
| 2744472 | -3.368 | protein binding, catalytic activity, translation initiation factor activity, binding | regulation of translational initiation, metabolism | BTB/POZ fold; Eukaryotic initiation factor 3, gamma subunit | |
| 2595644 | -3.455 | helicase activity, nucleic acid binding, ATP dependent helicase activity, ATP binding | | DNA/RNA helicase | |
| 2589748 | -3.549 | protein binding | | BAR; SWIB/MDM2 | |
| 2466521 | -3.599 | | | | |
| 2627153 | -3.831 | DNA binding, DNA-directed RNA polymerase activity | transcription | RNA polymerase, subunit H/Rpb5 C-terminal | Information storage and processing |
| 2515217 | -4.009 | translation initiation factor activity | translational initiation | Eukaryotic initiation factor 5A hypusine (eIF-5A) | |
| 2676065 | -4.249 | protein methyltransferase activity, iron ion binding, oxidoreductase activity | protein amino acid methylation | Isopenicillin N synthase; Ribosomal L11 methyltransferase | |
| 2679972 | -4.817 | DNA binding, protein binding, transcription factor activity | regulation of transcription, DNA-dependent, regulation of transcription | Beta-trefoil; LAG1, DNA binding; Beta-trefoil LAG1, DNA binding | |
| 2725570 | -4.835 | | | | |
| 2664466 | -4.922 | catalytic activity, nucleic acid binding, binding, oxidoreductase activity | metabolism | RNA recognition motif, RNP-1; Short-chain dehydrogenase/reductase SDR; Glucose/ribitol dehydrogenase | |
| 2677609 | -4.922 | polynucleotide adenylyltransferase activity, nucleotidyltransferase activity, RNA binding | transcription | Poly(A) polymerase, RNA-binding region; Nucleotidyltransferase, class I | |
| 2491982 | -5.006 | structural constituent of ribosome, RNA binding | protein biosynthesis | Ribosomal protein L23 | |
| 2692039 | -5.309 | proline-tRNA ligase activity, ATP binding, nucleotide binding, tRNA ligase activity | prolyl-tRNA aminoacylation, amino acid activation, protein biosynthesis | Prolyl-tRNA synthetase, class Iia | |
| 2734114 | -5.316 | helicase activity, nucleic acid binding, ATP dependent helicase activity, ATP binding | | DNA/RNA helicase | |

| | | | | | |
|---|---|---|---|---|---|
| 2674177 | -5.701 | translation release factor activity, codon specific | translational termination | Peptide chain release factor eRF/aRF subunit 1 | |
| 2743884 | -5.735 | protein kinase activity, transition metal ion binding, protein-tyrosine kinase activity, protein serine/threonine kinase activity, ATP binding, oxidoreductase activity | protein amino acid phosphorylation | Serine/threonine protein kinase-related; Tyrosine protein kinase; Serine/threonine protein kinase; Protein kinase ATP binding, conserved site | |
| 2488899 | -5.844 | structural constituent of ribosome | protein biosynthesis | Ribosomal protein S27e | |
| 2699460 | -5.867 | helicase activity, RNA binding, nucleic acid binding, ribonuclease III activity, ATP dependent helicase activity, ATP binding | RNA processing | DNA/RNA helicase; Ribonuclease III | |
| 2678425 | -6.187 | protein binding,GTP binding, GTPase activity | nucleocytoplasmic transport, signal transduction, DNA repair, small GTPase mediated signal transduction, protein transport, intracellular protein transport | Ras small GTPase, Ras type; Ras small GTPase, Rho type; Ras small GTPase, Rab type; Ran GTPase; Small GTP-binding protein | |
| 2675907 | -6.280 | | | | |
| 2599076 | -6.428 | protein targeting, intracellular protein transport | | Protein import receptor MAS20 | |
| 2601895 | -7.840 | calcium ion binding, ATP binding | mismatch repair | Calcium-binding EF-hand; DNA mismatch repair protein; ATP-binding region, ATPase-like | |
| 2680688 | -2.002 | phosphorylase activity, pyridoxal phosphate binding | carbohydrate metabolism | Glycosyl transferase, family 35; Glycogen/starch/alpha-glucan phosphorylase | |
| 2663700 | -2.021 | catalytic activity, binding, oxidoreductase activity, zinc ion binding | metabolism | Quinone oxidoreductase/zeta-crystallin, conserved site | |
| 2674539 | -2.053 | hydroxymethylglutaryl-CoA reductase (NADPH) activity, oxo-acid-lyase activity, anthranilate synthase activity, coenzyme binding | coenzyme A metabolism, biosynthesis, tryptophan biosynthesis | Hydroxymethylglutaryl-CoA reductase, class I/II; Anthranilate synthase component I | |
| 2600518 | -2.057 | transporter activity | transport | Sulphate transporter | |
| 2707128 | -2.087 | catalytic activity, DNA binding | DNA methylation, metabolism | C-5 cytosine-specific DNA methylase; Ribulose-phosphate binding barrel | |
| 2470775 | -2.126 | serine carboxypeptidase activity | proteolysis and peptidolysis | Peptidase S10, serine carboxypeptidase | |
| 2561893 | -2.132 | catalytic activity, transketolase activity | metabolism | Transketolase | |
| 2202445 | -2.140 | hydrogen-transporting ATPase activity, rotational mechanism, hydrogen ion transporter activity, hydrogen-transporting ATP synthase activity, rotational mechanism, hydrogen exporting ATPase activity, | energy coupled proton transport, against the electrochemical gradient, ATP synthesis coupled proton transport | ATPase, F1/V1/A1 complex | |

| | | | | | |
|---|---|---|---|---|---|
| | | phosphorylative mechanism | | | |
| 2752733 | -2.141 | catalytic activity, dTDP-4-dehydrorhamnose reductase activity, binding | extracellular polysaccharide biosynthesis, metabolism | dTDP-4-dehydrorhamnose reductase | |
| 2597019 | -2.152 | structural constituent of ribosome, acyl-CoA thioesterase activity | acyl-CoA metabolism, protein biosynthesis | Ribosomal protein, zinc-binding; Acyl-CoA thioesterase | |
| 2703628 | -2.152 | monooxygenase activity, heme binding, iron ion binding | electron transport | Cytochrome P450 | Metabolism |
| 2597721 | -2.152 | oxidoreductase activity, acting on NADH or NADPH | electron transport | ETC complex I subunit conserved region | |
| 2476486 | -2.171 | 3-isopropylmalate dehydratase activity | metabolism, leucine biosynthesis | 3-isopropylmalate dehydratase; Aconytase | |
| 2596037 | -2.175 | hydrogen-transporting ATPase activity, rotational mechanism, hydrogen ion transporter activity, nucleoside triphosphatase activity, nucleotide binding, hydrogen-transporting ATP, synthase activity, rotational mechanism, hydrogen-exporting ATPase activity, phosphorylative mechanism | ATP synthesis coupled proton transport, ATP biosynthesis | ATPase, F1/V1/A1 complex; AAA+ ATPase | |
| 2480368 | -2.183 | metal ion binding, superoxide dismutase activity | superoxide metabolism | Manganese and iron superoxide dismutase | |
| 2707033 | -2.212 | oxidoreductase activity | | Aldo/keto reductase | |
| 2254211 | -2.212 | glutamate-ammonia ligase activity | glutamine biosynthesis, nitrogen metabolism | Glutamine synthetase | |
| 2201427 | -2.231 | oxidoreductase activity, electron carrier activity | electron transport, tricarboxylic acid cycle | Alpha-helical ferredoxin; Succinate dehydrogenase/fumarate reductase iron-sulphur protein | |
| 2677337 | -2.231 | catalase activity | electron transport, response to oxidative stress | Catalase | |
| 2598890 | -2.236 | catalytic activity, binding, ubiquitin-like-protein ligase activity, glucose-6-phosphate 1-dehydrogenase activity | glucose metabolism, metabolism | Ubiquitin-conjugating enzyme, E2; Glucose-6-phosphate dehydrogenase | |
| 2437600 | -2.239 | hydrogen-transporting ATPase activity, rotational mechanism, hydrogen ion transporter activity, hydrogen-transporting ATP synthase activity, rotational mechanism, hydrogen exporting ATPase activity, phosphorylative mechanism | ATP synthesis coupled proton transport | ATPase, F1/V1/A1 complex | |

| | | | | | |
|---|---|---|---|---|---|
| 2676740 | -2.247 | hydrogen-transporting ATPase activity, rotational mechanism, hydrogen ion transporter activity, hydrogen-transporting ATP synthase activity, rotational mechanism, hydrolase activity, acting on acid anhydrides, catalyzing transmembrane movement of substances | ATP synthesis coupled proton transport | ATPase, F1/V1/A1 complex | |
| 2683366 | -2.260 | catalytic activity | metabolism | Crotonase, core | |
| 2599088 | -2.261 | phosphoadenylyl-sulfate reductase (thioredoxin) activity, RNA binding, ribonuclease III activity, transferase activity | RNA processing, sulfate assimilation, phosphoadenylyl sulfate reduction by phosphoadenylyl-sulfate reductase (thioredoxin) cysteine biosynthesis metabolism | Phosphoadenosine phosphosulphate reductase CysH-type; Ribonuclease III | |
| 2664848 | -2.262 | transaldolase activity | pentose-phosphate shunt, carbohydrate metabolism | Transaldolase AB | |
| 2692094 | -2.299 | histidinol-phosphate transaminase activity, transferase activity, transferring nitrogenous groups, transferase activity, pyridoxal phosphate binding | biosynthesis, metabolism, histidine biosynthesis | Histidinol-phosphate aminotransferase | |
| 2594260 | -2.306 | guanyl nucleotide binding, signal transducer activity | signal transduction, G-protein coupled receptor protein signaling pathway | Guanine nucleotide binding protein (G-protein), alpha subunit | |
| 2681988 | -2.309 | catalytic activity, malate dehydrogenase activity, binding, L-malate dehydrogenase activity, oxidoreductase activity | carbohydrate metabolism, TCA intermediate metabolism, malate metabolism, metabolism | Lactate dehydrogenase/glycoside hydrolase, family 4; Malate dehydrogenase, active site | |
| 2603431 | -2.311 | oxidoreductase activity | metabolism | Aldehyde dehydrogenase | |
| 2749900 | -2.365 | catalytic activity, inositol-3-phosphate synthase activity, binding | phospholipid biosynthesis, myo-inositol biosynthesis, metabolism | Myo-inositol-1-phosphate synthase | |
| 2681810 | -2.383 | hydrolase activity, oxidoreductase activity | | Metallophosphoesterase; Aldo-keto reductase | |
| 2599489 | -2.395 | catalytic activity, binding, coenzyme binding, DNA binding | metabolism | NAD-dependent epimerase/dehydratase | |
| 2744685 | -2.429 | hydrolase activity, acting on carbon-nitrogen (but not peptide) bonds | nitrogen metabolism | Nitrilase/cyanide hydratase and apolipoprotein N-acyltransferase | |

| | | | | | |
|---|---|---|---|---|---|
| 2701053 | -2.446 | | | | |
| 2674424 | -2.463 | catalytic activity, binding, oxidoreductase activity | metabolism | Short-chain dehydrogenase/reductase SDR; Glucose/ribitol dehydrogenase | |
| 2536606 | -2.465 | DNA binding, pantoate-beta-alanine ligase activity, DNA-directed RNA polymerase activity | transcription, pantothenate biosynthesis | Pantoate-beta-alanine ligase | |
| 2600459 | -2.476 | catalytic activity | trehalose biosynthesis | Glycosyl transferase, family 20; Trehalose-phosphatase | |
| 2623304 | -2.477 | chitinase activity, hydrolase activity, hydrolyzing O-glycosyl compounds | chitin catabolism, carbohydrate metabolism | Chitinase II; Glycoside hydrolase, family 18, catalytic domain | |
| 2504219 | -2.479 | guanyl nucleotide binding, GTP binding, signal transducer activity | signal transduction, G-protein coupled receptor protein signaling pathway | Fungal G-protein, alpha subunit | |
| 2295738 | -2.527 | metal ion transporter activity, zinc ion transporter activity | zinc ion transport, metal ion transport | Zinc/iron permease | |
| 2661549 | -2.529 | ubiquinol-cytochrome-c reductase activity | mitochondrial electron transport, ubiquinol to cytochrome c, electron transport | Cytochrome bd ubiquinol oxidase, 14 kDa subunit | |
| 2540747 | -2.530 | catalytic activity, FAD binding, oxidoreductase activity | FAD linked oxidase; Berberine/berberine-like | | |
| 2501050 | -2.533 | 1-pyrroline-5-carboxylate dehydrogenase activity, oxidoreductase activity | proline biosynthesis, metabolism | Delta-1-pyrroline-5-carboxylate dehydrogenase 1; Aldehyde dehydrogenase | |
| 2706145 | -2.534 | catalytic activity | | Hydantoinase B/oxoprolinase | |
| 2254325 | -2.575 | oxidoreductase activity, acting on the CH-OH group of donors, NAD or NADP as acceptor, isocitrate dehydrogenase (NAD) activity | metabolism, tricarboxylic acid cycle | Isocitrate/isopropylmalate dehydrogenase | Metabolism |
| 2597024 | -2.611 | catalytic activity, ATP binding | metabolism | ATP-citrate lyase/succinyl-CoA ligase; Succinyl-CoA synthetase, beta subunit; ATP-grasp fold | |
| 2674538 | -2.651 | catalytic activity, hydrolase activity, acting on ester bonds, binding, oxidoreductase activity | lipid metabolism, metabolism | Glucose/ribitol dehydrogenase; Short-chain dehydrogenase/reductase SDR; Lipase | |
| 2600574 | -2.694 | nucleobase transporter activity, glycogen (starch) synthase activity | glycogen biosynthesis, nucleobase, nucleoside, nucleotide and nucleic acid transport | Permease for cytosine/purines, uracil, thiamine, allantoin; Glycogen synthase | |
| 2598213 | -2.714 | catalytic activity, sulfite reductase (NADPH) activity, iron ion binding, electron carrier activity, NADPH binding, heme binding, oxidoreductase activity | electron transport, amino acid biosynthesis, metabolism | Transketolase, C-terminal/Pyruvate-ferredoxin oxidoreductase, domain II; Sulphite reductase; Flavodoxin-like | |

| | | | | | |
|---|---|---|---|---|---|
| 2672942 | -2.731 | carbohydrate binding, catalytic activity, isomerase activity | carbohydrate metabolism | Aldose 1-epimerase; Glycoside hydrolase-type carbohydrate-binding | |
| 2548345 | -2.732 | catalytic activity, phosphoribosylformylglycinamidine synthase activity | de novo' IMP biosynthesis | AIR synthase related protein; PurM, N-terminal-like; Phosphoribosylformylglycinamidine synthase, eukaryotes and proteobacteria | |
| 2598358 | -2.752 | catalytic activity, phosphoribosylaminoimidazolecarboxamide formyltransferase activity, IMP cyclohydrolase activity | IMP biosynthesis, purine nucleotide biosynthesis | AICARFT/IMPCHase bienzyme | |
| 2603451 | -2.755 | FAD binding, oxidoreductase activity | | Pyridine nucleotide-disulphide oxidoreductase, NAD-binding region | |
| 2674496 | -2.802 | catalytic activity, pyrroline-5-carboxylate reductase activity, binding | electron transport, proline biosynthesis, metabolism | Delta 1-pyrroline-5-carboxylate reductase | |
| 2273629 | -2.805 | cell communication | Phox-like | | |
| 2611460 | -2.823 | calcium ion binding, transporter activity, binding | transport | Calcium-binding EF-hand; Adenine nucleotide translocator 1; Mitochondrial carrier protein | Metabolism |
| 2108363 | -2.847 | catalytic activity, calcium ion binding, alpha,alpha-trehalase activity | trehalose catabolism, trehalose metabolism | Six-hairpin glycosidase-like; Glycoside hydrolase, family 37; Neutral trehalase Ca2+ binding | |
| 2625960 | -2.868 | citrate (Si)-synthase activity, transferase activity, transferring acyl groups, acyl groups converted into alkyl on transfer | | Citrate synthase, eukaryotic | |
| 2092683 | -2.964 | catalytic activity | metabolism, trehalose biosynthesis | Glycosyl transferase, family 20; Trehalose-phosphatase; HAD-superfamily hydrolase, subfamily IIB | |
| 2600967 | -2.995 | catalytic activity, oxidoreductase activity, acting on the CH-CH group of donors, binding, oxidoreductase activity | lipid metabolism, metabolism | 3-oxo-5-alpha-steroid 4-dehydrogenase; Short-chain dehydrogenase/reductase SDR; Glucose/ribitol dehydrogenase | |
| 2276847 | -3.015 | oxidoreductase activity | electron transport | NADH dehydrogenase (ubiquinone), 24 kDa subunit | |
| 2595597 | -3.040 | protein kinase activity, protein-tyrosine kinase activity, protein serine/threonine kinase activity, ATP binding | protein amino acid phosphorylation | Protein kinase; Serine/threonine protein kinase-related; Tyrosine protein kinase; Serine/threonine protein kinase | |
| 2598394 | -3.060 | structural constituent of ribosome, nucleic acid binding, methyltransferase activity | protein biosynthesis | Ribosomal protein L37ae | |
| 2748159 | -3.080 | oxidoreductase activity, acting on the CH-CH group of donors, transition metal ion binding, heme binding, acyl-CoA dehydrogenase activity | electron transport, metabolism | Acyl-CoA dehydrogenase/oxidase; Cythocrome b5 | |
| 2642095 | -3.081 | triose-phosphate isomerase activity | metabolism | Triosephosphate isomerase | Metabolism |

| | | | | | |
|---|---|---|---|---|---|
| 2585498 | -3.084 | ATP binding, diphosphomevalonate decarboxylase activity, kinase activity | isoprenoid biosynthesis, phosphorylation | GHMP kinase; Diphosphomevalonate decarboxylase | |
| 2751395 | -3.149 | catalytic activity, succinate-CoA ligase (ADP-forming) activity, binding, zinc ion binding, ATP citrate synthase activity, transferase activity, transferring acyl groups, acyl groups converted into alkyl on transfer | metabolism | ATP-citrate lyase/succinyl-CoA ligase | |
| 2679519 | -3.160 | catalytic activity, dihydroxy-acid dehydratase activity | branched chain family amino acid biosynthesis, metabolism | Dihydroxy-acid and 6-phosphogluconate dehydratase | |
| 2683261 | -3.264 | electron-transferring-flavoprotein dehydrogenase activity, oxidoreductase activity | electron transport | Electron transfer flavoprotein-ubiquinone oxidoreductase; Alpha-helical ferredoxin | |
| 2131899 | -3.304 | ATP binding | anion transport | Anion-transporting ATPase | |
| 2662238 | -3.326 | adenylosuccinate synthase activity, GTP binding | purine nucleotide biosynthesis | Adenylosuccinate synthetase | |
| 2056817 | -3.330 | oxidoreductase activity | metabolism | Aldehyde dehydrogenase | |
| 2695856 | -3.338 | | | | |
| 2695493 | -3.367 | oxidoreductase activity | carbohydrate metabolism, metabolism | Aldehyde dehydrogenase; Carbohydrate kinase, FGGY | |
| 2293651 | -3.382 | | | | |
| 2676205 | -3.390 | transporter activity, oxidoreductase activity, acting on the aldehyde or oxo group of donors, disulfide as acceptor, oxoglutarate dehydrogenase (lipoamide) activity | glycolysis, transport, metabolism | Major facilitator superfamily; Dehydrogenase, E1 component | |
| 2164570 | -3.394 | gamma-glutamyltransferase activity | | Gamma-glutamyltranspeptidase | |
| 2598986 | -3.455 | catalytic activity, binding, oxidoreductase activity, zinc ion binding | regulation of transcription, DNA-dependent, metabolism | Alcohol dehydrogenase, zinc-containing, conserved site | |
| 2676916 | -3.463 | binding, 3,4 dihydroxy-2-butanone-4-phosphate synthase activity | vitamin B2 biosynthesis, transport | Mitochondrial substrate carrier; 3,4-Dihydroxy-2-butanone 4-phosphate synthase | |
| 2599260 | -3.480 | oxidoreductase activity, acting on the CH-OH group of donors, NAD or NADP as acceptor, 3-isopropylmalate dehydrogenase activity | metabolism, leucine biosynthesis | Isocitrate/isopropylmalate dehydrogenase; 3-isopropylmalate dehydrogenase | |
| 2615440 | -3.508 | catalytic activity, binding, coenzyme binding | metabolism | NAD-dependent epimerase/dehydratase | |
| 2674964 | -3.516 | catalytic activity, protein kinase activity, protein serine/threonine kinase activity, ATP binding | protein amino acid phosphorylation, metabolism | Fumarylacetoacetase; Protein kinase; Serine/threonine protein kinase | |
| 2591856 | -3.537 | transporter activity | transport | Major facilitator superfamily | |

| | | | | | |
|---|---|---|---|---|---|
| 2146082 | -3.539 | oxidoreductase activity, acting on the CH-OH group of donors, NAD or NADP as acceptor, isocitrate dehydrogenase (NAD) activity | metabolism, tricarboxylic acid cycle | Isocitrate/isopropylmalate dehydrogenase | |
| 2116057 | -3.748 | acid phosphatase activity | | Histidine acid phosphatase | |
| 2676596 | -3.768 | catalytic activity, carboxyl- and carbamoyltransferase activity, monooxygenase activity, binding, amino acid binding, oxidoreductase activity | electron transport, amino acid metabolism, aromatic compound metabolism, metabolism | Aspartate/ornithine carbamoyltransferase; Monooxygenase, FAD-binding; Glucose/ribitol dehydrogenase; Pyridine nucleotide-disulphide oxidoreductase, class I; Aromatic-ring hydroxylase; Short-chain dehydrogenase/reductase SDR | |
| 2600373 | -3.830 | oxidoreductase activity, acting on sulfur group of donors, disulfide as acceptor, transcription factor activity | regulation of transcription, DNA-dependent, protein metabolism | Methionine sulphoxide reductase A; Homeobox | |
| 2597083 | -3.841 | acyltransferase activity | metabolism | Phospholipid/glycerol acyltransferase | |
| 2675304 | -3.843 | beta-galactosidase activity | carbohydrate metabolism | Glycoside hydrolase, family 35 | |
| 2675904 | -3.885 | hydrolase activity, hydrolyzing O-glycosyl compounds, oxidoreductase activity | carbohydrate metabolism | Glycoside hydrolase family 2, immunoglobulin-like beta-sandwich; Aldo/keto reductase | |
| 2582001 | -3.894 | hydrolase activity | | Hydantoinase/oxoprolinase | |
| 2282069 | -3.922 | oxidoreductase activity | metabolism | MaoC-like dehydratase | |
| 2750499 | -3.924 | catalytic activity, pyridoxal phosphate binding | cysteine biosynthesis from serine, metabolism | Pyridoxal phosphate-dependent enzyme, beta subunit; Cysteine synthase/cystathionine beta-synthase P-phosphate-binding site | |
| 2063627 | -3.941 | oxidoreductase activity | | FAD dependent oxidoreductase | |
| 2675512 | -3.963 | catalytic activity, transferase activity, transferring nitrogenous groups, transaminase activity, pyridoxal phosphate binding | biosynthesis, amino acid metabolism | Aspartate/other aminotransferase | |
| 2598507 | -4.027 | catalytic activity, oxidoreductase activity, acting on the CH-OH group of donors, NAD or NADP as acceptor, binding, oxidoreductase activity, zinc ion, inding | metabolism | Alcohol dehydrogenase, zinc-containing, conserved site; GroES-related | |
| 2685606 | -4.060 | ubiquitin-dependent protein catabolism | | Cullin | |
| 2597073 | -4.078 | amino acid transport, transport | | Amino acid permease, conserved site | |
| 2436819 | -4.080 | chromatin binding, binding, ATP binding, nucleotide binding, nucleoside triphosphatase activity, ATPase activity | chromatin assembly/disassembly | Chromo domain; Armadillo-type fold; ABC transporter-like; AAA+ ATPase | |
| 2595545 | -4.114 | methionine adenosyltransferase activity, ATP binding | one-carbon compound metabolism | S-adenosylmethionine synthetase | |
| 2679247 | -4.119 | catalytic activity, binding, oxidoreductase activity | fatty acid metabolism, metabolism | 3-hydroxyacyl-CoA dehydrogenase, NAD binding; 6-phosphogluconate dehydrogenase | |
| 2158355 | -4.197 | | | | |

| | | | | | |
|---|---|---|---|---|---|
| 2595683 | -4.201 | | L-serine biosynthesis, metabolism | Phosphoserine aminotransferase; Aminotransferase, class V/Cysteine desulphurase | |
| 2624619 | -4.274 | D-amino-acid oxidase activity, oxidoreductase activity | | D-amino acid oxidase, conserved site; FAD dependent oxidoreductase | |
| 2070913 | -4.294 | catalytic activity | carbohydrate metabolism | Glycosyl hydrolase, family 13, catalytic region | |
| 2567541 | -4.324 | catalytic activity, isocitrate lyase activity | carboxylic acid metabolism, metabolism | Isocitrate lyase and phosphorylmutase; Pyruvate/Phosphoenolpyruvate kinase, catalytic core | |
| 2700111 | -4.332 | catalytic activity | metabolism | Transketolase | |
| 2491930 | -4.356 | | | | |
| 2603678 | -4.367 | nucleoside triphosphatase activity, nucleotide binding, ATPase activity, ATP binding | | AAA+ ATPase, core; ABC transporter-like | |
| 2669280 | -4.420 | transporter activity, binding | transport | Adenine nucleotide translocator 1; Mitochondrial carrier protein | |
| 2562773 | -4.448 | beta-N-acetylhexosaminidase activity | carbohydrate metabolism | Glycoside hydrolase, family 20 | |
| 2680902 | -4.600 | carboxy-lyase activity, pyridoxal phosphate binding | carboxylic acid metabolism | Pyridoxal phosphate-dependent decarboxylase | |
| 2544123 | -4.624 | catalytic activity, GDP-mannose 4,6-dehydratase activity, binding, coenzyme binding, oxidoreductase activity | GDP-mannose metabolism, metabolism | NAD-dependent epimerase/dehydratase; GDP-mannose 4,6-dehydratase; Short-chain dehydrogenase/reductase SDR | |
| 2599939 | -4.644 | catalytic activity, magnesium ion binding | | Thiamine pyrophosphate enzyme, C-terminal TPP-binding | |
| 2679037 | -4.652 | GTP binding, oxidoreductase activity | cell cycle | Cell division/GTP binding protein; Redoxin | |
| 2535945 | -4.656 | phosphoribosylaminoimidazolesuccinocarboxamide synthase activity | purine nucleotide biosynthesis | SAICAR synthetase | |
| 2502569 | -4.678 | glycerophosphodiester phosphodiesterase activity | glycerol metabolism | Glycerophosphoryl diester phosphodiesterase | |
| 2338336 | -4.709 | catalytic activity | metabolism | Isocitrate lyase and phosphorylmutase; Pyruvate/Phosphoenolpyruvate kinase, catalytic core | |
| 2674764 | -4.720 | CoA-transferase activity | metabolism | Coenzyme A transferase; 3-oxoacid CoA-transferase, subunit B | |
| 2530104 | -4.838 | oxidoreductase activity | | NADH:flavin oxidoreductase/NADH oxidase | |
| 2473140 | -4.848 | catalytic activity, calcium ion binding, alpha-amylase activity | carbohydrate metabolism | Glycosyl hydrolase, family 13, subfamily, catalytic region; Alpha-amylase, fungi | |
| 2602081 | -4.866 | catalytic activity, binding | metabolism | NAD(P)-binding | |
| 2695732 | -4.870 | transferase activity, transferring nitrogenous groups, pyridoxal phosphate binding | biosynthesis | Aminotransferase, class I and II | |
| 2680115 | -4.980 | catalytic activity | | UBA/THIF-type NAD/FAD binding fold | |
| 2508662 | -5.077 | cysteine-type endopeptidase activity | proteolysis and peptidolysis | Peptidase C1B, bleomycin hydrolase | |
| 2312113 | -5.224 | catalytic activity, binding, coenzyme binding | metabolism | NAD-dependent epimerase/dehydratase | |
| 2512669 | -5.251 | catalytic activity, adenosylhomocysteinase activity, binding | one-carbon compound metabolism, metabolism | S-adenosyl-L-homocysteine hydrolase | |

| | | | | | |
|---|---|---|---|---|---|
| 2551610 | -5.268 | peroxidase activity, ATP binding, nucleoside triphosphatase activity, nucleotide binding, heme binding, ATPase activity, ATPase activity, coupled to transmembrane movement of substances | electron transport, transport, response to oxidative stress | ABC transporter, transmembrane region; AAA+ ATPase; ABC transporter-like | |
| 2704408 | -5.285 | glucose-6-phosphate isomerase activity | glycolysis, gluconeogenesis | Phosphoglucose isomerase (PGI) | |
| 2511932 | -5.338 | catalytic activity, binding, oxidoreductase activity | electron transport, metabolism | Oxidoreductase | |
| 2596034 | -5.454 | | regulation of signal transduction, response to biotic stimulus | TAP42-like protein | |
| 2750135 | -5.490 | glucosamine-6-phosphate deaminase activity | N-acetylglucosamine metabolism, carbohydrate metabolism | Glucosamine-6-phosphate isomerase | |
| 2026034 | -5.531 | protein kinase activity, protein-tyrosine kinase activity, protein serine/threonine kinase activity, ATP binding | protein amino acid phosphorylation | Protein kinase; Serine/threonine protein kinase-related; Protein kinase ATP binding, conserved site | |
| 2676329 | -5.613 | catalytic activity, phospho-N-acetylmuramoyl-pentapeptide-transferase activity, isocitrate lyase activity | carboxylic acid metabolism, metabolism | Isocitrate lyase and phosphorylmutase; Pyruvate/Phosphoenolpyruvate kinase, catalytic core; Glycosyl transferase, family 4 | |
| 2706632 | -5.648 | carbohydrate binding, chitinase activity, hydrolase activity, hydrolyzing O-glycosyl compounds | chitin catabolism, carbohydrate metabolism | Carbohydrate-binding family V/XII; Chitinase II; Glycoside hydrolase, family 18, catalytic domain | |
| 2552020 | -5.672 | catalytic activity | acetyl-CoA metabolism | Acetyl-CoA hydrolase/transferase | |
| 2681688 | -5.715 | transferase activity, transferring nitrogenous groups, pyridoxal phosphate binding | biosynthesis | Aminotransferase, class I and II | |
| 2274225 | -6.028 | metal ion binding, superoxide dismutase activity | superoxide metabolism | Manganese and iron superoxide dismutase | |
| 2255769 | -6.240 | | | Translationally controlled tumour-associated TCTP | |
| 2549499 | -6.499 | catalytic activity, alpha,alpha-trehalose-phosphate synthase (UDP-forming) activity | trehalose biosynthesis | Glycosyl transferase, family 20; Alpha,alpha-trehalose-phosphate synthase | |
| 2592701 | -6.581 | catalytic activity, oxidoreductase activity, acting on CH-OH group of donors, binding, oxidoreductase activity, acting on the CH-OH group of donors, NAD or ADP as acceptor glycerol-3-phosphate dehydrogenase (NAD) activity oxidoreductase activity | glycerol-3-phosphate catabolism, glycerol-3-phosphate metabolism, carbohydrate metabolism, metabolism | NAD-dependent glycerol-3-phosphate dehydrogenase | |
| 2469166 | -6.720 | | | | |

| | | | | | |
|---|---|---|---|---|---|
| 1152047 | -6.904 | catalytic activity | | Epoxide hydrolase-like | |
| 2637783 | -7.508 | catalytic activity, carboxy-lyase activity, magnesium ion binding | | Thiamine pyrophosphate enzyme, central region; Pyruvate decarboxylase/indolepyruvate decarboxylase | Metabolism |
| 2714417 | -8.756 | | | | |
| 2753281 | -2.118 | metalloexopeptidase activity | proteolysis and peptidolysis | Peptidase M24, catalytic core | |
| 2502834 | -2.134 | hydrolase activity | | Dienelactone hydrolase | |
| 2596562 | -2.136 | catalytic activity, binding, phosphogluconate dehydrogenase (decarboxylating) activity, sugar porter activity, transporter activity, oxidoreductase activity | carbohydrate transport, transport, pentose-phosphate shunt, metabolism | 6-phosphogluconate dehydrogenase; General substrate transporter; Major facilitator superfamily | |
| 2675965 | -2.149 | catalytic activity | metabolism | Type I phosphodiesterase/nucleotide pyrophosphatase/phosphate transferase; Alkaline-phosphatase-like, core domain | |
| 2242809 | -2.164 | binding | | Armadillo-type fold | |
| 2680350 | -2.214 | transporter activity | transport | Major facilitator superfamily | |
| 2597208 | -2.264 | GTP binding, phosphopyruvate hydratase activity | glycolysis | GTP1/OBG; GTP-binding protein, HSR1-related; Enolase | |
| 2198127 | -2.381 | | | | |
| 2596927 | -2.427 | nucleic acid binding, ATP binding, helicase activity, sugar porter activity, transporter activity, ATP dependent helicase activity | carbohydrate transport, transport | DNA/RNA helicase; Sugar transporter | |
| 2678297 | -2.571 | GTP binding | | GTP-binding protein, HSR1-related; GTP1/OBG | |
| 2599835 | -2.578 | catalytic activity, binding, carbohydrate binding, transcriptional repressor activity | carbohydrate catabolism, regulation of nitrogen utilization, metabolism | Carbohydrate-binding family 9/cellobiose dehydrogenase, cytochrome | |
| 2495704 | -2.595 | catalytic activity, epoxide hydrolase activity | response to toxin | Epoxide hydrolase-like | |
| 2597423 | -2.822 | hydrogen ion transporter activity, hydrogen-transporting ATPase activity, rotational mechanism, hydrogen-transporting ATP synthase activity, rotational mechanism | ATP synthesis coupled proton transport | ATPase, F1/V1/A1 complex | |
| 2532844 | -2.822 | | | Adhesion regulating molecule | |
| 2687919 | -2.839 | catalytic activity | metabolism | Amidohydrolase 2 | |
| 2565304 | -2.913 | catalytic activity | | Esterase/lipase/thioesterase | |
| 2749685 | -2.983 | sugar porter activity, transporter activity | carbohydrate transport, transport | General substrate transporter; Major facilitator superfamily; Sugar transporter, conserved site | |
| 2596102 | -3.021 | catalytic activity, structural constituent of ribosome, magnesium ion binding, phospholipid-translocating ATPase activity, ATP binding, ATPase activity, coupled to transmembrane | phospholipid transport, protein biosynthesis, transport, metabolism | Haloacid dehalogenase-like hydrolase; Ribosomal protein S2; Phospholipid-translocating P-type ATPase, flippase; ATPase, P-type, K/Mg/Cd/Cu/Zn/Na/Ca/Na/H-transporter | |

| | | | | | |
|---|---|---|---|---|---|
| | | movement of ions, phosphorylative mechanism | | | |
| 2680499 | -3.096 | cytochrome-c oxidase activity | electron transport | Cytochrome c oxidase, subunit Vib | |
| 2693747 | -3.313 | transporter activity, binding | signal transduction, transport | Rho GTPase activation protein; Lipocalin | |
| 2593151 | -3.319 | | | | |
| 2677686 | -3.331 | transferase activity, transferring nitrogenous groups, GTPase activity, GTP binding, structural constituent of nuclear pore, pyridoxal phosphate binding | biosynthesis | Aminotransferase, class I and II; Dynamin GTPase effector; Nucleoporin, Nsp1-like | |
| 2608284 | -3.340 | carboxypeptidase A activity, chaperone activator activity, ATPase stimulator activity, zinc ion binding | proteolysis and peptidolysis | Peptidase M14; Activator of Hsp90 ATPase | |
| 2677972 | -3.389 | structural constituent of ribosome, GTP binding | protein transport, small GTPase mediated signal transduction, protein biosynthesis | Ribosomal protein S7e; Ras small GTPase, Ras type; Ras small GTPase, Rho type; Ras small GTPase, Rab type; Small GTP-binding protein | |
| 2679553 | -3.479 | N-acetyltransferase activity | metabolism | GCN5-related N-acetyltransferase | |
| 2746421 | -3.525 | GTP binding | protein transport, small GTPase mediated signal transduction | Ras small GTPase, Ras type; Ras small GTPase, Rho type; Ras small GTPase, Rab type; Small GTP-binding protein | |
| 2596033 | -3.568 | FAD binding, oxidoreductase activity, acting on CH-OH group of donors, GTP binding | protein transport, small GTPase mediated signal transduction | Glucose-methanol-choline oxidoreductase; Ras GTPase; Ras small GTPase, Ras type; Ras small GTPase, Rab type | |
| 2231044 | -3.629 | | | | |
| 2595487 | -3.663 | nucleotide binding | | tRNA-binding arm | |
| 2501262 | -3.810 | lipid metabolism, metabolism | | Patatin; Acyl transferase/acyl hydrolase/lysophospholipase | |
| 2674907 | -3.813 | catalytic activity, GTP binding, magnesium ion binding, phospholipid-translocating ATPase activity, ATP binding, ATPase activity, coupled to transmembrane movement of ions, phosphorylative mechanism | cell cycle, phospholipid transport, transport, metabolism | Cell division/GTP binding protein; Phospholipid-translocating P-type ATPase, flippase; ATPase, P-type, K/Mg/Cd/Cu/Zn/Na/Ca/Na/H-transporter; Phospholipid-translocating P-type ATPase, flippase | |
| 2242729 | -3.961 | | | | |
| 2674209 | -4.182 | GTPase activity, GTP binding | | Dynamin, GTPase region | |
| 2675760 | -4.210 | hydrolase activity | | Dienelactone hydrolase | |
| 2669543 | -4.425 | GTP binding | protein transport, small GTPase mediated signal transduction | Ras small GTPase, Ras type; Ras small GTPase, Rho type; Ras small GTPase, Rab type; Small GTP-binding protein | |
| 2681776 | -4.459 | aminopeptidase activity | proteolysis and peptidolysis | Peptidase M17, leucyl aminopeptidase | |
| 2254126 | -4.723 | | | | |

| | | | | | |
|---|---|---|---|---|---|
| 2696773 | -4.743 | | | | |
| 2660812 | -4.925 | oxidoreductase activity | negative regulation of transcription | Flavodoxin/nitric oxide synthase | |
| 2674809 | -4.927 | phosphoric ester hydrolase activity, nucleic acid binding, zinc ion binding | carbohydrate metabolism | Inositol phosphatase/fructose-1,6-bisphosphatase | |
| 2676133 | -4.970 | FAD binding, oxidoreductase activity, acting on CH-OH group of donors | | Glucose-methanol-choline oxidoreductase | |
| 2643023 | -5.165 | FAD binding, oxidoreductase activity, acting on CH-OH group of donors | | Glucose-methanol-choline oxidoreductase | |
| 2604988 | -5.310 | endocytosis | | Adaptin ear-binding coat-associated protein 1 NECAP-1 | |
| 2676541 | -5.552 | | | | |
| 2696064 | -5.791 | FAD binding, oxidoreductase activity, acting on CH-OH group of donors | alcohol metabolism | Glucose-methanol-choline oxidoreductase | |
| 2601683 | -5.845 | catalytic activity | metabolism | Haloacid dehalogenase-like hydrolase | |
| 2706358 | -5.909 | catalytic activity | metabolism | Haloacid dehalogenase-like hydrolase | |
| 2688236 | -6.243 | catalytic activity, binding, ATP binding, nucleotide binding, tRNA ligase activity, oxidoreductase activity | amino acid activation, protein biosynthesis, metabolism | Aminoacyl-tRNA synthetase, class I, conserved site; Short-chain dehydrogenase/reductase SDR; Glucose/ribitol dehydrogenase | |
| 2676989 | -6.511 | cysteine protease inhibitor activity | | Proteinase inhibitor I25, cystatin | |
| 2600127 | -7.567 | | | | |
| 2619664 | -2.002 | | | | |
| 2681715 | -2.031 | transporter activity, motor activity, ATP binding | transport | Major facilitator superfamily; Myosin head | |
| 2676395 | -2.033 | aspartic-type endopeptidase activity, pepsin A activity | proteolysis and peptidolysis | Peptidase aspartic; Peptidase A1 | |
| 2703675 | -2.036 | | | | |
| 2596743 | -2.062 | translation initiation factor activity, RNA binding | translational initiation | Eukaryotic translation initiation factor 4E (eIF-4E) | |
| 2592761 | -2.078 | | | | |
| 2749597 | -2.082 | transporter activity | transport | Xanthine/uracil/vitamin C permease | |
| 2563170 | -2.097 | | | | |
| 2675614 | -2.098 | metal ion transporter activity | metal ion transport | Zinc/iron permease | |
| 2586510 | -2.140 | hydrolase activity, acting on carbon-nitrogen (but not peptide) bonds | | Metal-dependent hydrolase, composite | |
| 2678251 | -2.147 | DNA binding, cyanate hydratase activity, hydro-lyase activity | cyanate metabolism, regulation of transcription, DNA-dependent, metabolism | High mobility group box, HMG1/HMG2; Lambda repressor-like, DNA-binding; Cyanase | |
| 2596872 | -2.155 | | | | |
| 2629150 | -2.205 | | | | |
| 2663278 | -2.215 | tRNA binding | tRNA-binding region | | |

| | | | | | |
|---|---|---|---|---|---|
| 2597807 | -2.226 | ubiquitin-like-protein ligase activity | | Ubiquitin-conjugating enzyme, E2 | |
| 2678034 | -2.234 | | | Mpv17/PMP22 | |
| 2630944 | -2.260 | | | | |
| 2600430 | -2.341 | mRNA metabolism | | Like-Sm ribonucleoprotein | |
| 2747968 | -2.387 | | | | |
| 2601836 | -2.394 | protein binding | | Leucine-rich repeat | |
| 2674849 | -2.399 | catalytic activity, 2-isopropylmalate synthase activity, O-methyltransferase activity, transferase activity, transferring acyl groups, acyl groups converted into alkyl on transfer | carboxylic acid metabolism, leucine biosynthesis | Pyruvate carboxyltransferase; 2-isopropylmalate synthase LeuA; Yeast 2-isopropylmalate synthase; O-methyltransferase, family 2 | |
| 2677861 | -2.399 | catalytic activity | | Six-hairpin glycosidase-like | |
| 1126744 | -2.427 | | | | |
| 2599659 | -2.428 | phosphoenolpyruvate carboxykinase (ATP) activity, phosphoenolpyruvate carboxykinase activity, ATP binding, purine nucleotide binding | gluconeogenesis | Phosphoenolpyruvate carboxykinase, ATP-utilising | |
| 2510342 | -2.482 | protein binding | cell communication | Phox-like | |
| 2681191 | -2.486 | catalytic activity, hydrolase activity, acting on carbon-nitrogen (but not peptide) bonds, hydrolase activity | metabolism | Haloacid dehalogenase-like hydrolase; Metal-dependent hydrolase, composite; Amidohydrolase | |
| 2740159 | -2.489 | carbohydrate binding, hydrolase activity, hydrolyzing O-glycosyl compounds | carbohydrate metabolism | Carbohydrate-binding family V/XII | |
| 2673553 | -2.490 | catalytic activity, hydrolase activity, acting on carbon-nitrogen (but not peptide) bonds | carbohydrate metabolism | Polysaccharide deacetylase; Glycoside hydrolase/deacetylase, beta/alpha-barrel | |
| 2512880 | -2.561 | | | | |
| 2484388 | -2.564 | | | | |
| 2679777 | -2.586 | phosphoenolpyruvate carboxykinase (ATP) activity, phosphoenolpyruvate carboxykinase activity, ATP binding, purine nucleotide binding | gluconeogenesis | Phosphoenolpyruvate carboxykinase, ATP-utilising | |
| 2508404 | -2.637 | | | | |
| 2674721 | -2.641 | calcium ion binding | | Calcium-binding EF-hand | |
| 2517922 | -2.693 | binding | | Armadillo-type fold | |
| 2601132 | -2.738 | hydrolase activity, hydrolyzing O-glycosyl compounds, beta-lactamase activity, zinc ion binding | antibiotic catabolism, carbohydrate metabolism | Glycoside hydrolase, family 43; Beta-lactamase, class B, conserved site | |
| 2599032 | -2.748 | nucleoside triphosphatase activity, nucleotide binding, ATPase activity, coupled to transmembrane movement of substances, ATPase activity, ATP binding | transport | AAA+ ATPase, core; ABC transporter, transmembrane region | |

| | | | | | |
|---|---|---|---|---|---|
| 2599051 | -2.778 | nucleic acid binding, integrase activity | mitochondrial genome maintenance | Integrase C; Mitochondrial genome maintenance MGM101 | |
| 2608339 | -2.786 | | | | |
| 2532725 | -2.801 | oxidoreductase activity, acting on paired donors, with oxidation of a pair of donors resulting in the reduction of molecular oxygen to two molecules of water, oxidoreductase activity | lipid metabolism | Fatty acid desaturase, type 1 | |
| 2538728 | -2.808 | ribonuclease activity, RNA binding | | Ribonuclease II and R | |
| 2677721 | -2.856 | nucleic acid binding, zinc ion binding, DNA binding, transcription factor activity, pyridoxal phosphate binding | amino acid metabolism, regulation of transcription, DNA-dependent, transcription | RNA recognition motif, RNP-1; Fungal transcriptional regulatory protein; Cys/Met metabolism, pyridoxal phosphate-dependent enzyme | |
| 2675838 | -2.925 | asparagine synthase (glutamine-hydrolyzing) activity, nucleic acid binding | asparagine biosynthesis | Asparagine synthase; RNA recognition, region 1; Prohibitin | |
| 2600933 | -2.929 | beta-1,4-mannosylglycoprotein 4-beta-N-acetylglucosaminyltransferase activity | N-linked glycosylation | Glycosyl transferase, family 17 | |
| 2597110 | -2.955 | | | | |
| 2677111 | -2.988 | | | | |
| 2600273 | -3.041 | catalytic activity, structural constituent of ribosome, binding, oxidoreductase activity | protein biosynthesis, transport, metabolism | Ribosomal protein L27e; Short-chain dehydrogenase/reductase SDR; Glucose/ribitol dehydrogenase | |
| 2600484 | -3.054 | | | | |
| 2555894 | -3.058 | | | | |
| 2621933 | -3.105 | nucleic acid binding | | Polynucleotidyl transferase, Ribonuclease H fold | |
| 2702757 | -3.132 | protein binding | | BTB/POZ fold | |
| 2747169 | -3.171 | catalytic activity, aspartate-semialdehyde dehydrogenase activity, binding, zinc ion binding, NADPH binding, protein dimerization activity, oxidoreductase activity, acting on the aldehyde or oxo group of donors, NAD or NADP as acceptor, transcription factor activity | amino acid metabolism, methionine biosynthesis, amino acid biosynthesis, threonine biosynthesis, metabolism, regulation of transcription, DNA-dependent | Fungal transcriptional regulatory protein; Aspartate-semialdehyde dehydrogenase | |
| 2680985 | -3.174 | catalytic activity, FAD binding, oxidoreductase activity | | FAD-binding, type 2 | |
| 2597620 | -3.176 | | | | |
| 1186194 | -3.181 | | | | |
| 2703645 | -3.199 | | | | |
| 2696916 | -3.210 | O-methyltransferase activity | | O-methyltransferase, family 2 | |
| 2597062 | -3.221 | mannose-1-phosphate guanylyltransferase (GDP) activity, | polysaccharide metabolism, biosynthesis | Mannose-6-phosphate isomerase, type II, C-terminal | |

| | | | | | |
|---|---|---|---|---|---|
| | | nucleotidyltransferase activity | | | |
| 2580701 | -3.258 | two-component sensor molecule activity, protein-histidine kinase activity, two-component response regulator activity, signal transducer activity, ATP binding, transferase activity, transferring phosphorus-containing groups | signal transduction, phosphorylation, regulation of transcription, DNA-dependent, two-component signal transduction system (phosphorelay), peptidyl-histidine phosphorylation | Signal transduction histidine kinase | |
| 2677212 | -3.271 | catalytic activity | metabolism | Haloacid dehalogenase-like hydrolase | |
| 2676276 | -3.271 | 3-deoxy-7-phosphoheptulonate synthase activity, binding | biosynthesis, transport, aromatic amino acid family biosynthesis | DHAP synthase, class 1; Mitochondrial substrate carrier | |
| 2677275 | -3.285 | | | | |
| 2528625 | -3.351 | | | | |
| 2579864 | -3.411 | | | | |
| 2601385 | -3.419 | binding | transport | Mitochondrial substrate carrier | |
| 2595924 | -3.426 | | | | |
| 2142247 | -3.515 | RNA binding, binding | | Pumilio RNA-binding region; Armadillo-type fold | |
| 2476052 | -3.550 | | | | |
| 2676356 | -3.658 | catalytic activity, thiosulfate sulfurtransferase activity, chorismate mutase activity, carbon-carbon lyase activity | cell redox homeostasis, aromatic compound metabolism, sulfate transport, aromatic amino acid family biosynthesis | Pyruvate/Phosphoenolpyruvate kinase, catalytic core; Thiosulphate sulphurtransferase, conserved site; Chorismate mutase | |
| 2567631 | -3.720 | | | | Information storage and processing |
| 2639294 | -3.745 | protein modification | | Ubiquitin | |
| 2546333 | -3.751 | protein binding | | BTB/POZ-like | |
| 2567746 | -3.784 | | | | |
| 2471655 | -3.790 | | | | |
| 2198994 | -3.800 | | | | |
| 2117151 | -3.874 | | | | |
| 2195418 | -3.876 | | | | |
| 2585457 | -3.901 | peroxidase activity, heme binding | electron transport, response to oxidative stress | Haem peroxidase | |
| 2565612 | -3.918 | | | | |
| 2708788 | -3.920 | protein kinase activity, protein-tyrosine kinase activity, protein serine/threonine kinase activity, ATP binding | protein amino acid phosphorylation | Protein kinase; Serine/threonine protein kinase-related; Tyrosine protein kinase | |

| | | | | | |
|---|---|---|---|---|---|
| 2678233 | -3.927 | catalytic activity, binding, oxidoreductase activity, ketol-acid reductoisomerase activity | branched chain family amino acid biosynthesis, metabolism | 6-phosphogluconate dehydrogenase; Acetohydroxy acid isomeroreductase | |
| 2463002 | -3.936 | | | | |
| 2681284 | -3.952 | catalytic activity | nucleoside metabolism | Nucleoside phosphorylase | |
| 2675792 | -4.044 | catalytic activity, histone deacetylase activity | histone deacetylation | Histone deacetylase | |
| 2596805 | -4.227 | motor activity, binding, ATP binding | | Myosin head, motor region | |
| 2081064 | -4.236 | phosphoric monoester hydrolase activity | metabolism | HAD-superfamily hydrolase, subfamily IB, PSPase-like | |
| 2600338 | -4.244 | | | | |
| 2599803 | -4.256 | glutamate decarboxylase activity, carboxy-lyase activity, transcription factor activity, pyridoxal phosphate binding | carboxylic acid metabolism, glutamate metabolism, regulation of transcription, DNA-dependent | Pyridoxal phosphate-dependent decarboxylase; Glutamate decarboxylase; Homeobox | |
| 2677843 | -4.268 | prenyltransferase activity, nucleic acid binding, Rho GDP-dissociation inhibitor activity, zinc ion binding | | RHO protein GDP dissociation inhibitor; UbiA prenyltransferase | |
| 2601175 | -4.270 | zinc ion binding, pepsin A activity, DNA binding, aspartic-type endopeptidase activity, transcription factor activity | regulation of transcription, DNA-dependent, proteolysis and peptidolysis, transcription | Fungal specific transcription factor; Peptidase A1 | |
| 2746951 | -4.271 | | | | |
| 2548110 | -4.285 | | | | |
| 2601160 | -4.366 | | | | |
| 2565384 | -4.378 | catalytic activity, binding, oxidoreductase activity | metabolism | 6-phosphogluconate dehydrogenase | |
| 2264463 | -4.385 | hydrolase activity, hydrolyzing O-glycosyl compounds | carbohydrate metabolism | Glycoside hydrolase, family 17 | |
| 2539960 | -4.525 | hydrolase activity, hydrolyzing O-glycosyl compounds | carbohydrate metabolism | Glycoside hydrolase, family 3 | |
| 2500167 | -4.553 | nucleoside triphosphatase activity, nucleotide binding, ATPase activity, ATP binding | | AAA+ ATPase; ABC transporter-like | |
| 2456797 | -4.578 | 1-aminocyclopropane-1-carboxylate deaminase activity, catalytic activity, pyridoxal phosphate binding | metabolism, amine catabolism | 1-aminocyclopropane-1-carboxylate deaminase; Pyridoxal phosphate-dependent enzyme, beta subunit | |
| 2526241 | -4.625 | binding | | Armadillo-type fold | |
| 2336543 | -4.696 | | | Legume-like lectin | |
| 2753486 | -4.771 | | | | |
| 2753424 | -4.818 | | | | |
| 2689252 | -4.852 | nucleotide binding, tRNA ligase activity, ATP binding, oxidoreductase activity | amino acid activation, protein biosynthesis | Aminoacyl-tRNA synthetase, class I, conserved site | |
| 2680457 | -4.960 | zinc ion binding | | Zinc finger, MYND-type | |

| | | | | | |
|---|---|---|---|---|---|
| 2283081 | -4.983 | | metabolism | Aminotransferase, class V/Cysteine desulphurase | |
| 2597310 | -4.983 | protein binding, peptidyl-prolyl cis-trans isomerase activity | protein folding | Peptidyl-prolyl cis-trans isomerase, cyclophilin-type; BTB/POZ-like | |
| 2500013 | -5.003 | transition metal ion binding, motor activity, heme binding, ATP binding, transferase activity, transferring hexosyl groups | | Myosin head, motor region; Cythochrome b5 | |
| 2599368 | -5.061 | | | Peptidase S54, rhomboid | |
| 2678193 | -5.083 | monooxygenase activity, oxidoreductase activity | electron transport, aromatic compound metabolism, metabolism | Monooxygenase, FAD-binding; Aromatic-ring hydroxylase | |
| 2600547 | -5.127 | catalytic activity, carboxyl- and carbamoyltransferase activity, ATP binding, aspartate carbamoyltransferase activity, carbamoyl-phosphate synthase, amino acid binding | glutamine metabolism, amino acid metabolism, biosynthesis, metabolism, 'de novo' pyrimidine base biosynthesis, nitrogen metabolism | Glutamine amidotransferase superfamily; Anthranilate synthase component II/delta crystallin; Carbamoyl phosphate synthase, GATase region | |
| 2508802 | -5.180 | | | | |
| 2601164 | -5.182 | structural constituent of ribosome, FAD binding, oxidoreductase activity, ureidoglycolate hydrolase activity, protein binding, ATP binding, DNA binding, allantoicase activity | protein biosynthesis, electron transport, regulation of transcription, DNA-dependent, intra-Golgi transport, allantoin catabolism | Ribosomal protein S9; Pyridine nucleotide-disulphide oxidoreductase, NAD-binding region; Ureidoglycolate hydrolase; SMCs flexible hinge | |
| 2540154 | -5.215 | | | | |
| 2672812 | -5.224 | | | | |
| 2574569 | -5.670 | | | | |
| 2460130 | -5.758 | | | | |
| 2452507 | -5.759 | | | | |
| 2103552 | -5.826 | signal transducer activity | | Regulator of G protein signalling | |
| 2574999 | -5.905 | catalytic activity, protein kinase activity, ATP binding | protein amino acid phosphorylation | Six-hairpin glycosidase-like; Protein kinase ATP binding, conserved site | |
| 2595248 | -5.949 | serine carboxypeptidase activity | proteolysis and peptidolysis | Peptidase S10, serine carboxypeptidase | |
| 2742326 | -6.085 | | | | |
| 2535816 | -6.140 | | | | |
| 2597177 | -6.211 | isomerase activity, protein binding, oxidoreductase activity | electron transport | Peptidyl-prolyl cis-trans isomerase, PpiC-type | |
| 2439795 | -6.488 | | | | |
| 2620107 | -6.504 | | | | |
| 2537084 | -6.678 | | | | |
| 2602110 | -6.691 | | | | |
| 2678455 | -6.805 | | | | |

| | | | | | |
|---|---|---|---|---|---|
| 2595350 | -6.962 | GTP binding | protein transport, small GTPase mediated signal transduction | Ras small GTPase, Ras type; Ras small GTPase, Rho type; Ras small GTPase, Rab type; Small GTP-binding protein | |
| 2640541 | -6.974 | | | | |
| 2184213 | -7.160 | | | | |
| 1138359 | -7.751 | | | | |
| 2594802 | -8.521 | | | | |

### Table S22. Repressed proteins in fruiting bodies vs. monokaryon 4-39

| Schco3 ID | Fold change | Go Desc molecular function | Go Desc Biological Process | Interpro Desc | KOG Group |
|---|---|---|---|---|---|
| 2227218 | -6.866 | nucleoside triphosphatase activity, nucleotide binding, hydrolase activity, ATP binding | protein catabolism | AAA+ ATPase, core; 26S proteasome subunit P45; AAA ATPase, conserved site | |
| 2566045 | -2.053 | catalytic activity, metalloendopeptidase activity, metal ion binding, zinc ion binding | proteolysis and peptidolysis | Metalloenzyme, LuxS/M16 peptidase-like, metal-binding; Peptidase M16 | |
| 2601828 | -2.054 | | | | |
| 2616372 | -2.237 | protein transporter activity, binding | protein-nucleus import, docking, intracellular protein transport | | cytoplasm, nuclear pore, nucleus |
| 2565354 | -2.261 | RAB GDP-dissociation inhibitor activity | protein transport | Rab GDI protein | |
| 2597001 | -2.299 | | | | |
| 2606323 | -2.313 | biosynthesis | | Glycosyl transferase, group 1 | Cellular processes and signaling |
| 2601695 | -2.336 | | | | |
| 2721824 | -2.385 | protein binding | cell wall organization and biogenesis, protein localization | Pili assembly chaperone; Vacuolar protein sorting-associated protein | |
| 2599910 | -2.388 | protein binding | | Actin/actin-like | |
| 2694313 | -2.420 | | | | |
| 2555639 | -2.429 | | | | |
| 2481091 | -2.504 | | | | |
| 2458589 | -2.509 | | | | |
| 2056522 | -2.537 | DNA binding | | Histone-fold | |
| 2595575 | -2.553 | GTP binding | small GTPase mediated signal transduction, intracellular protein transport | ARF/SAR superfamily, ADP-ribosylation factor, GTP-binding protein SAR1, Small GTP-binding protein | |
| 2594767 | -2.555 | protein binding, ATP binding | | Actin/actin-like | |
| 2679029 | -2.558 | protein kinase CK2, intrinsic regulator activity, DNA binding | protein folding | Casein kinase II, regulatory subunit; Histone-fold; Heat shock protein DnaJ; Transcription factor CBF/NF-Y/archaeal histone | |
| 2175961 | -2.580 | protein kinase activity, MAP kinase activity, protein-tyrosine kinase activity, protein | protein amino acid phosphorylation | Protein kinase; Serine/threonine protein kinase-related; Protein kinase ATP binding, conserved site; MAP kinase, conserved site | |

| | | | | | |
|---|---|---|---|---|---|
| | | serine/threonine kinase activity, ATP binding | | | |
| 2601822 | -2.581 | catalytic activity, zinc ion binding | | Six-hairpin glycosidase-like; Zinc finger, MYND-type | |
| 2599395 | -2.584 | catalytic activity, small protein activating enzyme activity | protein modification, ubiquitin cycle | UBA/THIF-type NAD/FAD binding fold; Ubiquitin-activating enzyme, E1 | |
| 2532770 | -2.608 | regulation of actin polymerization | | ARP2/3 complex 16 kDa subunit (p16-Arc) | |
| 2746768 | -2.611 | protein binding | cell communication | Phox-like | |
| 2528872 | -2.625 | protein kinase activity, protein-tyrosine kinase activity, protein serine/threonine kinase activity, ATP binding | protein amino acid phosphorylation | Protein kinase ATP binding, conserved site; Serine/threonine protein kinase, active site; Tyrosine protein kinase | |
| 2600987 | -2.647 | endopeptidase activity, shikimate kinase activity, ATP binding, threonine endopeptidase activity, kinase activity | ubiquitin-dependent protein catabolism, carbohydrate metabolism | Proteasome alpha-subunit, conserved site; 20S proteasome, A and B subunits; Carbohydrate kinase, thermoresistant glucokinase; Shikimate kinase | |
| 2709241 | -2.655 | ATP binding | protein folding | ATP-binding region, ATPase-like; Heat shock protein Hsp90 | |
| 2603783 | -2.669 | transition metal ion binding, heme binding, binding | | Cytochrome b5; Armadillo-type fold | |
| 2678491 | -2.760 | guanyl-nucleotide exchange factor activity, isomerase activity, small GTPase regulatory/interacting protein activity, Rho guanyl-nucleotide exchange factor activity | intracellular signaling cascade, cell redox homeostasis, regulation of Rho protein signal transduction | Guanine-nucleotide dissociation stimulator, CDC24, conserved site; Disulphide isomerase; Citron-like; Dbl homology (DH) domain | |
| 2603003 | -2.783 | ubiquitin-like-protein ligase activity | | Ubiquitin-conjugating enzyme, E2 | |
| 2271670 | -2.799 | endopeptidase activity, threonine endopeptidase activity | ubiquitin-dependent protein catabolism | 20S proteasome, A and B subunits; Proteasome alpha-subunit, conserved site | |
| 2601975 | -2.803 | | | | |
| 2533584 | -2.862 | serine-type peptidase activity | proteolysis and peptidolysis | Peptidase S9, prolyl oligopeptidase active site region | |
| 2488292 | -2.873 | | | | |
| 2188670 | -2.905 | catalytic activity, metalloendopeptidase activity, metal ion binding, zinc ion binding | proteolysis and peptidolysis | Metalloenzyme, LuxS/M16 peptidase-like, metal-binding; Peptidase M16 | |
| 2674818 | -2.911 | catalytic activity, formate-tetrahydrofolate ligase activity, binding, 1,3-beta-glucan synthase activity, ATP binding | beta-1,3 glucan biosynthesis, folic acid and derivative biosynthesis, metabolism | Tetrahydrofolate dehydrogenase/cyclohydrolase; NAD(P)-binding; Formate-tetrahydrofolate ligase, FTHFS | |
| 2458250 | -2.987 | peptidase activity | proteolysis and peptidolysis | Peptidase M28 | |
| 2453944 | -3.027 | phosphoric monoester, hydrolase, activity, protein tyrosine/serine/threonine phosphatase activity, protein tyrosine phosphatase activity | protein amino acid dephosphorylation, dephosphorylation | Protein-tyrosine phosphatase | |

| | | | | | |
|---|---|---|---|---|---|
| 2349132 | -3.041 | protein kinase activity, protein-tyrosine kinase activity, protein serine/threonine kinase activity, ATP binding | protein amino acid phosphorylation | Protein kinase, core; Serine/threonine protein kinase-related; Protein kinase ATP binding, conserved site; Tyrosine protein kinase | |
| 2600008 | -3.088 | protein binding, structural molecule activity, binding, transporter activity, protein transporter activity | protein complex assembly, transport, intracellular protein transport, vesicle-mediated transport | Clathrin/coatomer adaptor, adaptin-like; Coatomer, beta subunit; Armadillo-type fold; Major facilitator superfamily | |
| 2599999 | -3.130 | catalytic activity, binding, protein transporter activity, importin-alpha export receptor activity | cell proliferation, protein-nucleus import, docking, apoptosis, intracellular protein transport | Phosphatidic acid phosphatase type 2/haloperoxidase; Armadillo-type fold, Importin-beta; CAS/CSE | |
| 2601777 | -3.150 | autophagy | | Autophagy protein 5 | |
| 2207599 | -3.182 | actin binding | | Actin-binding, cofilin/tropomyosin type | |
| 2331063 | -3.186 | protein binding, ATP binding | protein folding | Chaperonin Cpn60/TCP-1; GroEL-like chaperone, ATPase | |
| 2016319 | -3.252 | protein binding, GTP binding, GTPase activity | nucleocytoplasmic transport, signal transduction, small GTPase mediated signal transduction, protein transport, intracellular protein transport | Ran GTPase; Ras small GTPase, Ras type; Ras small GTPase, Rho type; Ras small GTPase, Rab type; Small GTP-binding protein | |
| 2492200 | -3.322 | serine-type peptidase activity | proteolysis and peptidolysis | Peptidase S28 | |
| 2161007 | -3.344 | protein binding, binding, protein transporter activity | protein complex assembly, intracellular protein transport, vesicle-mediated transport | Clathrin/coatomer adaptor, adaptin-like; Armadillo-type fold | |
| 2666833 | -3.353 | nucleoside metabolism | | Phosphoribosyltransferase | |
| 2599106 | -3.369 | nucleoside triphosphatase activity, nucleotide binding, serine-type peptidase activity, binding, ATP binding | | AAA+ ATPase, core; Peptidase S53, propeptide; Aspartate decarboxylase-like fold; 1 Cell division protein 48 | |
| 2676846 | -3.403 | protein binding, structural molecule activity,binding, protein transporter activity | protein complex assembly, intracellular protein transport, vesicle-mediated transport | Clathrin, heavy chain/VPS, 7-fold repeat | |
| 2596185 | -3.429 | protein binding, GTP binding, GTPase activity | nucleocytoplasmic transport, signal transduction, small GTPase mediated signal | Ras small GTPase, Ras type; Ras small GTPase, Rho type; Ras small GTPase, Rab type; Ran GTPase; Small GTP-binding protein | |

| | | | | | |
|---|---|---|---|---|---|
| | | | transduction, protein transport, intracellular protein transport | | |
| 2675504 | -3.459 | metalloexopeptidase activity, hydrolase activity, methionyl aminopeptidase activity | metabolism, proteolysis and peptidolysis | Alpha/beta hydrolase fold-3; Lipase, GDXG, active site; Peptidase M24 | |
| 2545132 | -3.505 | GTPase activity, structural molecule activity, GTP binding | microtubule-based movement, microtubule-based process | Tubulin/FtsZ, C-terminal; Beta tubulin | |
| 2276972 | -3.520 | | | | |
| 2531252 | -3.561 | UDP-glucose 4-epimerase activity, catalytic activity, binding, coenzyme binding | metabolism, galactose metabolism | UDP-glucose 4-epimerase | |
| 2249899 | -3.578 | dolichyl-diphosphooligosaccharide-protein glycosyltransferase activity | protein amino acid glycosylation | Ribophorin I | |
| 2603157 | -3.581 | aspartic-type endopeptidase activity, transporter activity, pepsin A activity | oligopeptide transport, proteolysis and peptidolysis | Peptidase aspartic, catalytic | |
| 2704716 | -3.599 | nucleoside triphosphatase activity, nucleotide binding, hydrolase activity, ATP binding | protein catabolism | AAA+ ATPase, core; 26S proteasome subunit P45 | |
| 1157296 | -3.606 | ATP binding | | Heat shock protein Hsp70 | |
| 2709667 | -3.616 | protein binding, motor activity, ATP binding | | Myosin S1 fragment; Myosin head | |
| 2249278 | -3.625 | | | | |
| 2597970 | -3.628 | aspartic-type endopeptidase activity, pepsin A activity | proteolysis and peptidolysis | Peptidase aspartic, catalytic; Peptidase A1 | |
| 2602808 | -3.637 | protein prenyltransferase activity | protein amino acid prenylation | Protein prenyltransferase, alpha subunit | |
| 2678444 | -3.653 | protein kinase activity, protein-tyrosine kinase activity, protein serine/threonine kinase activity, ATP binding, N-acetyltransferase activity | protein amino acid phosphorylation, metabolism | Protein kinase, core; Serine/threonine protein kinase-related; Tyrosine protein kinase; Serine/threonine protein kinase | |
| 2535557 | -3.697 | regulation of actin polymerization | | Arp2/3 complex, 34kDa subunit p34-Arc | |
| 2574819 | -3.709 | | | | |
| 2378688 | -3.761 | | | | |
| 2601514 | -3.770 | protein binding, ATP binding | protein folding | Chaperonin Cpn60/TCP-1; T-complex protein 1, zeta subunit | |
| 2608113 | -3.773 | catalytic activity, metalloendopeptidase activity, metal ion binding, zinc ion binding | proteolysis and peptidolysis | Metalloenzyme, LuxS/M16 peptidase-like, metal-binding | |
| 2596581 | -3.854 | protein binding | cell communication | Phox-like | |
| 2266989 | -3.895 | | | | |
| 2464773 | -3.905 | protein binding | | Actin/actin-like | |
| 2685426 | -3.998 | protein binding, ATP binding | protein folding | Chaperonin Cpn60/TCP-1; T-complex protein 1, gamma subunit; Chaperonin TCP-1, conserved site; GroEL-like chaperone, ATPase | |

| | | | | | |
|---|---|---|---|---|---|
| 2033700 | -4.004 | catalytic activity, small protein activating enzyme activity | protein modification, ubiquitin cycle | UBA/THIF-type NAD/FAD binding fold; Ubiquitin-activating enzyme, E1 | |
| 2481214 | -4.019 | carboxypeptidase A activity, zinc ion binding | proteolysis and peptidolysis | Peptidase M14, carboxypeptidase A | |
| 2597008 | -4.072 | protein folding | | Heat shock protein DnaJ | |
| 2674326 | -4.073 | microtubule motor activity, ATP binding | microtubule-based movement | Kinesin, motor region | |
| 2750934 | -4.083 | protein folding | | Peptidyl-prolyl cis-trans isomerase, FKBP-type | |
| 2706648 | -4.121 | protein binding, protein transporter activity | protein complex assembly, transport, vesicle-mediated transport, intracellular protein transport | Clathrin adaptor, mu subunit | |
| 2675835 | -4.157 | 6-phosphogluconolactonase activity | pentose-phosphate shunt, carbohydrate metabolism | 6-phosphogluconolactonase; Glucosamine/galactosamine-6-phosphate isomerase | |
| 2685615 | -4.174 | actin binding | actin cytoskeleton organization and biogenesis | F-actin capping protein, alpha subunit | |
| 2676856 | -4.207 | calcium ion binding, DNA binding, nucleotide-sugar transporter activity, oxidoreductase activity | electron transport, nucleotide-sugar transport | Calcium-binding EF-hand; SANT, DNA-binding; UDP-galactose transporter; Flavin-containing amine oxidase | |
| 2595504 | -4.217 | protein kinase activity, protein serine/threonine kinase activity, nucleic acid binding, ATP binding, protein-tyrosine kinase activity, ATPase activity, voltage-dependent ion-selective channel activity | protein amino acid phosphorylation, anion transport | Protein kinase; ABC transporter-like; Serine/threonine protein kinase-related; Tyrosine protein kinase; mitochondrial outer membrane | |
| 2674535 | -4.234 | protein binding, ATP binding | protein folding | Chaperonin Cpn60; T-complex protein 1; GroEL-like chaperone, ATPase | |
| 2701639 | -4.251 | threonine endopeptidase activity | ubiquitin-dependent protein catabolism | 20S proteasome, A and B subunits | |
| 2599215 | -4.264 | GTPase activity, structural molecule activity, GTP binding | microtubule-based movement, microtubule-based process | Alpha tubulin | |
| 2603876 | -4.267 | | | | |
| 2595604 | -4.358 | catalytic activity, Rab GTPase activator activity, binding, oxidoreductase activity | metabolism | RabGAP/TBC | |
| 2599607 | -4.376 | glutathione peroxidase activity | response to oxidative stress | Glutathione peroxidase | |
| 2597700 | -4.465 | nucleoside triphosphatase activity, nucleotide binding, hydrolase activity, ATP binding | protein catabolism | AAA ATPase; 26S proteasome subunit P45 | |
| 2544615 | -4.481 | peptidase activity, serine-type peptidase activity | signal peptide processing, | Peptidase S26B, eukaryotic signal peptidase | |

| | | | | | |
|---|---|---|---|---|---|
| | | | proteolysis and peptidolysis | | |
| 2498761 | -4.504 | | | | |
| 2482758 | -4.528 | nucleoside triphosphatase activity, nucleotide binding, hydrolase activity, ATP binding | protein catabolism | AAA ATPase; 26S proteasome subunit P45 | |
| 2600989 | -4.547 | ATP binding, binding, DNA binding, DNA topoisomerase type II activity | signal transduction, DNA metabolism, transport, DNA topological change | Spo11/DNA topoisomerase VI, subunit A; Mitochondrial substrate carrier; Rho GAP | |
| 2596663 | -4.648 | nucleotidyltransferase activity | metabolism | UTP--glucose-1-phosphate uridylyltransferase | |
| 2576552 | -4.692 | protein kinase activity, protein-tyrosine kinase activity, protein serine/threonine kinase activity, ATP binding | protein amino acid phosphorylation | Protein kinase; Serine/threonine protein kinase-related; Tyrosine protein kinase | |
| 2675738 | -4.778 | intracellular protein transport | | NSF attachment protein | |
| 2598895 | -4.799 | protein binding | cell communication | Phox-like | |
| 2521613 | -4.956 | nucleoside metabolism | | Phosphoribosyltransferase | |
| 2678401 | -5.043 | GTPase activity, structural molecule activity, GTP binding | microtubule-based movement, microtubule-based process | Beta tubulin | |
| 2678570 | -5.063 | protein binding, binding, peptidyl-prolyl cis-trans isomerase activity | protein folding, RNA metabolism | Peptidyl-prolyl cis-trans isomerase, cyclophilin-type; MIF4G-like, type 3; Armadillo-type fold | |
| 2681354 | -5.152 | protein binding; zinc ion binding | | Zinc finger, RING-type | |
| 2723169 | -5.156 | protein transporter activity, binding | protein-nucleus import, docking, intracellular protein transport | Importin-beta; Armadillo-type fold | |
| 2582680 | -5.250 | binding | | Armadillo-type fold | |
| 2602353 | -5.263 | | | | |
| 2754081 | -5.313 | antioxidant activity, oxidoreductase activity | Alkyl hydroperoxide reductase/ Thiol specific antioxidant/ Mal allergen | | |
| 2744020 | -5.342 | protein binding, GTP binding,GTPase activity | nucleocytoplasmic transport, signal transduction, small GTPase mediated signal transduction, protein transport, intracellular protein transport | Ras small GTPase, Ras type; Ras small GTPase, Rho type; Ras small GTPase, Rab type; Ran GTPase; Small GTP-binding protein | |
| 2041665 | -5.348 | | | | |
| 2490330 | -5.397 | | | | |
| 2480386 | -5.443 | protein binding, ATP binding | protein folding | Chaperonin Cpn60 | |
| 2602003 | -5.445 | protein binding, binding, ATP binding, RNA ligase (ATP) activity | tRNA splicing | DEATH-like; Armadillo-type fold; tRNA ligase, phosphodiesterase | |

| | | | | | |
|---|---|---|---|---|---|
| 2247557 | -5.628 | | | | |
| 2500485 | -5.981 | chitin synthase activity | chitin biosynthesis | Chitin synthase | |
| 2680097 | -6.460 | acid phosphatase activity, zinc ion binding, transcription factor activity, ARF guanyl-nucleotide exchange factor activity | regulation of transcription, DNA-dependent | Histidine acid phosphatase; SEC7-like | |
| 2699882 | -7.555 | nucleic acid binding, peptidyl-prolyl cis-trans isomerase activity | protein folding | RNA recognition motif, RNP-1; Peptidyl-prolyl cis-trans isomerase, cyclophilin-type | |
| 2677294 | -7.850 | catalytic activity, binding, coenzyme binding | metabolism | NAD-dependent epimerase/dehydratase | |
| 2675082 | -9.979 | DNA binding, DNA-directed RNA polymerase activity | transcription | DNA-directed RNA polymerase, M/15 kDa subunit | |
| 2599165 | -5.893 | transcription factor activity; threonine endopeptidase activity; zinc ion binding | ubiquitin-dependent protein catabolism, regulation of transcription, DNA-dependent | Fungal transcriptional regulatory protein, N-terminal; 20S proteasome, A and B subunits; | |
| 2271597 | -5.938 | intracellular transport | | Ran Binding Protein 1 | |
| 2622052 | -2.072 | nucleic acid binding | | Polynucleotidyl transferase, Ribonuclease H fold | |
| 2601442 | -2.100 | nucleic acid binding | response to stress | OsmC-like protein; Cleavage and polyadenylation specificity factor, A subunit | |
| 2676444 | -2.116 | | | | |
| 2675609 | -2.121 | catalytic activity, nucleic acid binding, binding, carbohydrate binding, DNA binding, isomerase activity, nucleotide binding, DNA-directed DNA polymerase activity | nucleobase, nucleoside, nucleotide and nucleic acid metabolism, carbohydrate metabolism, DNA replication | DNA-directed DNA polymerase, family B; Polynucleotidyl transferase, Ribonuclease H fold | |
| 2700057 | -2.131 | phenylalanine-tRNA ligase activity, ATP binding, nucleotide binding, transition metal ion binding, RNA binding, heme binding | phenylalanyl-tRNA aminoacylation, protein biosynthesis | B3/B4 tRNA-binding domain; Phenylalanyl-tRNA synthetase, class IIc, beta subunit, archae/euk cytosolic; Putative DNA binding | |
| 2635723 | -2.141 | structural constituent of ribosome | protein biosynthesis | Ribosomal protein L30e | Information storage and processing |
| 2599032 | -2.147 | nucleoside triphosphatase activity, nucleotide binding, ATPase activity, coupled to transmembrane movement of substances, ATPase activity, ATP binding | transport | AAA+ ATPase, core; ABC transporter, transmembrane region | |
| 2466521 | -2.301 | | | | |
| 2598506 | -2.346 | nucleic acid binding, ATP binding, DNA binding, helicase activity, transcription factor activity | DNA methylation, regulation of transcription, DNA-dependent, RNA splicing | DNA/RNA helicase; SNF2-related; C-5 cytosine-specific DNA methylase | |

| | | | | | |
|---|---|---|---|---|---|
| 2595892 | -2.351 | ATP binding, metal ion binding, zinc ion binding, nucleotide binding, hydrolase activity, acting on carbon-nitrogen (but not peptide) bonds, in linear amidines, tRNA ligase activity | amino acid activation, protein biosynthesis | Aminoacyl-tRNA synthetase, class I, conserved site; Ureohydrolase; Zinc finger, MYND-type | |
| 2644636 | -2.387 | structural constituent of ribosome | protein biosynthesis | Ribosomal protein L28e | |
| 2675703 | -2.424 | catalytic activity, succinate-CoA ligase (ADP-forming) activity, phosphotransferase activity, alcohol group as acceptor, binding, ATP citrate synthase activity, transferase activity, transferring phosphorus-containing groups, acyl-CoA binding | metabolism | Succinyl-CoA ligase, alpha subunit; Armadillo-type fold; Phosphatidylinositol 3- and 4-kinase | |
| 2681753 | -2.487 | ATP binding, nucleotide binding, histidine-tRNA ligase activity, tRNA ligase activity | histidyl-tRNA aminoacylation, amino acid activation, protein biosynthesis | Anticodon-binding; Histidyl-tRNA synthetase, class IIa, subgroup; Aminoacyl-tRNA synthetase, class II | |
| 2745550 | -2.513 | structural constituent of ribosome | protein biosynthesis | Ribosomal protein S19e | |
| 2662721 | -2.579 | transcription regulator activity | regulation of transcription | Basic helix-loop-helix dimerisation region bHLH; Helix-loop-helix DNA-binding | |
| 2595220 | -2.643 | farnesyl-diphosphate farnesyltransferase activity, transferase activity, zinc ion binding | biosynthesis, lipid biosynthesis | Farnesyl-diphosphate farnesyltransferase; Squalene/phytoene synthase; Zinc finger, MYND-type | |
| 2354702 | -2.752 | translation initiation factor activity | translational initiation | Eukaryotic translation initiation factor 3 subunit 8, N-terminal | |
| 2354737 | -2.778 | transferase activity | | Trimeric LpxA-like | |
| 2595644 | -2.839 | helicase activity, nucleic acid binding, ATP dependent helicase activity, ATP binding | | DNA/RNA helicase | |
| 2354342 | -2.868 | structural constituent of ribosome, nucleic acid binding, RNA binding | protein biosynthesis | Ribosomal protein S13 | |
| 2596209 | -2.870 | helicase activity, nucleic acid binding, ATP dependent helicase activity, ATP binding | | DNA/RNA helicase | |
| 2505976 | -2.910 | DNA binding, transcription regulator activity, A-directed RNA polymerase activity, nucleic acid binding, zinc ion binding | transcription, regulation of transcription | DNA-directed RNA polymerase, M/15 kDa subunit; Zinc finger, TFIIS-type | |
| 2705009 | -2.914 | leucine-tRNA ligase activity, ATP binding, nucleotide binding, tRNA ligase activity | leucyl-tRNA aminoacylation, amino acid activation, protein biosynthesis | Leucyl-tRNA synthetase, class Ia, archaeal/eukaryotic cytosolic; Aminoacyl-tRNA synthetase, class 1a, anticodon-binding; Valyl/Leucyl/Isoleucyl-tRNA synthetase, class Ia, editing | |
| 2599769 | -3.007 | protein binding, GTP binding, nucleic acid binding, ATP binding, zinc ion binding, helicase | small GTPase mediated signal transduction, protein transport | Ras small GTPase, Ras type; Ras small GTPase, Rho type; Ras small GTPase, Rab type; Small GTP-binding protein | |

| | | | | | |
|---|---|---|---|---|---|
| | | activity, ATP dependent helicase activity, | | | |
| 2598311 | -3.009 | argininosuccinate synthase activity, NAD-dependent histone deacetylase activity, ATP binding, zinc ion binding, DNA binding | arginine biosynthesis, regulation of transcription, DNA-dependent, protein amino acid deacetylation, chromatin silencing | Argininosuccinate synthase; NAD-dependent histone deacetylase, silent information regulator Sir2 | |
| 2557698 | -3.040 | structural constituent of ribosome | protein biosynthesis | Ribosomal protein L21e | |
| 2492699 | -3.075 | translation initiation factor activity, RNA binding | protein biosynthesis | Eukaryotic translation initiation factor 2, alpha subunit; S1, RNA binding | |
| 2623763 | -3.089 | | | SET | Information storage and processing |
| 2544254 | -3.106 | DNA binding, DNA polymerase processivity factor activity | regulation of DNA replication | Proliferating cell nuclear antigen, PCNA | |
| 2679031 | -3.187 | protein binding, nucleic acid binding, ATP binding, helicase activity, protein transporter activity, ATP dependent helicase activity | protein complex assembly, transport, intracellular protein transport, vesicle-mediated transport | Clathrin adaptor, mu subunit; DANN RNA Helicase | |
| 2749954 | -3.226 | structural constituent of ribosome | protein biosynthesis | Ribosomal protein S11 | |
| 2678650 | -3.227 | structural constituent of ribosome | protein biosynthesis | Ribosomal protein L31e | |
| 2147637 | -3.229 | aspartate-tRNA ligase activity, nucleic acid binding, ATP binding, nucleotide binding, transporter activity, tRNA ligase activity | amino acid activation, protein biosynthesis, aspartyl-tRNA aminoacylation, transport | Aspartyl-tRNA synthetase, class Iib | |
| 2548156 | -3.379 | alanine-tRNA ligase activity, ligase activity, forming aminoacyl-tRNA and related compounds, nucleic acid binding, ATP binding | alanyl-tRNA aminoacylation, protein biosynthesis | Alanyl-tRNA synthetase, class Iic; Threonyl/alanyl tRNA synthetase, SAD; Phosphoesterase, DHHA1 | |
| 2744472 | -3.504 | protein binding, catalytic activity, translation initiation factor activity, binding | regulation of translational initiation, metabolism | BTB/POZ fold; Eukaryotic initiation factor 3, gamma subunit | |
| 2598148 | -3.603 | structural constituent of ribosome, ATP binding, DNA helicase activity | protein biosynthesis | Ribosomal protein S19/S15; AAA ATPase, core; TIP49 | |
| 2627153 | -3.681 | DNA binding, DNA-directed RNA polymerase activity | transcription | RNA polymerase, subunit H/Rpb5 C-terminal | Information storage and processing |
| 2598258 | -3.698 | carbon-nitrogen ligase activity, with glutamine as amido-N-donor, binding | | Amidase signature enzyme; Armadillo-type fold | |
| 2749322 | -3.818 | structural constituent of ribosome | protein biosynthesis | Ribosomal protein S17e | |

| | | | | | |
|---|---|---|---|---|---|
| 2602338 | -3.923 | tRNA binding, tRNA-pseudouridine synthase I activity, nucleic acid binding, ATP binding, DNA binding, ATPase activity | pseudouridine synthesis, tRNA processing | tRNA pseudouridine synthase; DNA/RNA helicase, C-terminal; SNF2-related; ABC transporter-like | |
| 2681298 | -3.926 | thiamin diphosphokinase activity, ATP binding, nucleotide binding, lycine-tRNA ligase activity, tRNA ligase activity | thiamin diphosphate biosynthesis, amino acid activation, thiamin metabolism, protein biosynthesis, glycyl-tRNA aminoacylation | Thiamin pyrophosphokinase, catalytic region; Glycyl-tRNA synthetase, alpha2 dimer; Aminoacyl-tRNA synthetase, class II (G, H, P and S); Anticodon-binding | |
| 2491982 | -3.991 | structural constituent of ribosome, RNA binding | protein biosynthesis | Ribosomal protein L23 | |
| 2696887 | -4.084 | protein binding, DNA binding, transcription factor activity, oxidoreductase activity | electron transport, regulation of transcription, DNA-dependent | WW/Rsp5/WWP; Histone-fold; FMN-dependent alpha-hydroxy acid dehydrogenase; Bacterial regulatory protein, LuxR | |
| 2743884 | -4.165 | protein kinase activity, transition metal ion binding, protein-tyrosine kinase activity, protein serine/threonine kinase activity, ATP binding, oxidoreductase activity | protein amino acid phosphorylation | Serine/threonine protein kinase-related; Tyrosine protein kinase; Serine/threonine protein kinase; Protein kinase ATP binding, conserved site | |
| 2515217 | -4.175 | translation initiation factor activity | translational initiation | Eukaryotic initiation factor 5A hypusine (eIF-5A) | |
| 2166139 | -4.237 | structural constituent of ribosome | protein biosynthesis | Ribosomal protein S9 | |
| 2674545 | -4.262 | structural constituent of ribosome | protein biosynthesis | Ribosomal protein S2 | |
| 2680176 | -4.501 | structural constituent of ribosome | protein biosynthesis | Ribosomal protein L11 | |
| 2599066 | -4.588 | carbon-nitrogen ligase activity, with glutamine as amido-N-donor, ligase activity | protein biosynthesis | GatB, central region; GatB/Yqey; Glutamyl-tRNA(Gln) amidotransferase B subunit; GatB N-terminal region; Glutamyl-tRNA(Gln) amidotransferase B subunit | |
| 2599058 | -4.646 | translation initiation factor activity | translational initiation | Eukaryotic translation initiation factor 3, subunit 7 | |
| 2677609 | -4.708 | polynucleotide adenylyltransferase activity, nucleotidyltransferase activity, RNA binding | transcription | Poly(A) polymerase, RNA-binding region; Nucleotidyltransferase, class I | |
| 2692039 | -4.721 | proline-tRNA ligase activity, ATP binding, nucleotide binding, tRNA ligase activity | prolyl-tRNA aminoacylation, amino acid activation, protein biosynthesis | Prolyl-tRNA synthetase, class Iia | |
| 2595535 | -4.766 | GTPase activity, GTP binding, RNA binding | | Protein synthesis factor, GTP-binding; Translation elongation factor EFG/EF2; Translation elongation factor EFTu/EF1A, domain 2; Small GTP-binding protein; GTP-binding protein LepA; K Homology | |
| 2620095 | -4.776 | | | | |

| | | | | | |
|---|---|---|---|---|---|
| 2743097 | -4.867 | catalytic activity, binding | carbohydrate metabolism | Glycosyl hydrolase, family 13, catalytic region; Armadillo-type fold | |
| 1319169 | -4.998 | transition metal ion binding, heme binding, nucleic acid binding, zinc ion binding | | Cytochrome b5 | |
| 2451194 | -5.192 | DNA binding, zinc ion binding | | SANT, DNA-binding; Zinc finger ZZ type | |
| 2734114 | -5.217 | helicase activity, nucleic acid binding, ATP dependent helicase activity, ATP binding | | DNA/RNA helicase | |
| 2589748 | -5.235 | protein binding | | BAR; SWIB/MDM2 | |
| 2599076 | -5.359 | protein targeting, intracellular protein transport | | Protein import receptor MAS20 | |
| 2675907 | -5.469 | | | | |
| 2676065 | -5.553 | protein methyltransferase activity, iron ion binding, oxidoreductase activity | protein amino acid methylation | Isopenicillin N synthase; Ribosomal L11 methyltransferase | |
| 2488899 | -5.571 | structural constituent of ribosome | protein biosynthesis | Ribosomal protein S27e | |
| 2596995 | -5.926 | nucleic acid binding, ATP binding, DNA binding, helicase activity, hydrolase activity, hydrolyzing O-glycosyl compounds | carbohydrate metabolism | DNA/RNA helicase; SNF2-related | |
| 2746016 | -6.102 | protein binding | | Leucine-rich repeat | |
| 2679972 | -6.114 | DNA binding, protein binding, transcription factor activity | regulation of transcription, DNA-dependent, regulation of transcription | Beta-trefoil; LAG1, DNA binding; Beta-trefoil LAG1, DNA binding | |
| 2677971 | -6.218 | GTPase activity, GTP binding | | Protein synthesis factor, GTP-binding; Translation elongation factor EFTu/EF1A, domain 2 | |
| 2674177 | -6.309 | translation release factor activity, codon specific | translational termination | Peptide chain release factor eRF/aRF subunit 1 | |
| 2106720 | -6.353 | ribosome binding | mature ribosome assembly | Translation initiation factor IF6 | |
| 2525411 | -6.678 | rRNA binding | rRNA processing | Gar1 protein RNA-binding region | |
| 2595617 | -6.755 | structural constituent of ribosome | protein biosynthesis | Ribosomal protein S21e | |
| 2664466 | -6.827 | catalytic activity, nucleic acid binding, binding, oxidoreductase activity | metabolism | RNA recognition motif, RNP-1; Short-chain dehydrogenase/reductase SDR; Glucose/ribitol dehydrogenase | |
| 2678425 | -6.898 | protein binding,GTP binding, GTPase activity | nucleocytoplasmic transport, signal transduction, DNA repair, small GTPase mediated signal transduction, protein transport, intracellular protein transport | Ras small GTPase, Ras type; Ras small GTPase, Rho type; Ras small GTPase, Rab type; Ran GTPase; Small GTP-binding protein | |
| 2747126 | -6.977 | structural constituent of ribosome | protein biosynthesis | Ribosomal protein S21e | |

| | | | | | |
|---|---|---|---|---|---|
| 2601895 | -7.802 | calcium ion binding, ATP binding | mismatch repair | Calcium-binding EF-hand; DNA mismatch repair protein; ATP-binding region, ATPase-like | |
| 2748326 | -2.012 | | | | |
| 2697737 | -2.013 | catalytic activity, binding, oxidoreductase activity | amino acid metabolism, metabolism | Glutamate/phenylalanine/leucine/valine dehydrogenase | |
| 2594260 | -2.053 | guanyl nucleotide binding, signal transducer activity | signal transduction, G-protein coupled receptor protein signaling pathway | Guanine nucleotide binding protein (G-protein), alpha subunit | |
| 2108363 | -2.068 | catalytic activity, calcium ion binding, alpha,alpha-trehalase activity | trehalose catabolism, trehalose metabolism | Six-hairpin glycosidase-like; Glycoside hydrolase, family 37; Neutral trehalase Ca2+ binding | |
| 2254325 | -2.077 | oxidoreductase activity, acting on the CH-OH group of donors, NAD or NADP as acceptor, isocitrate dehydrogenase (NAD) activity | metabolism, tricarboxylic acid cycle | Isocitrate/isopropylmalate dehydrogenase | Metabolism |
| 2675512 | -2.087 | catalytic activity, transferase activity, transferring nitrogenous groups, transaminase activity, pyridoxal phosphate binding | biosynthesis, amino acid metabolism | Aminotransferases, class-I, pyridoxal-phosphate-binding site | |
| 2597874 | -2.103 | protein binding, ribonucleoside-diphosphate reductase activity | DNA replication | Ribonucleotide reductase R1 subunit, N-terminal | |
| 2585498 | -2.139 | ATP binding, diphosphomevalonate decarboxylase activity, kinase activity | isoprenoid biosynthesis, phosphorylation | GHMP kinase; Diphosphomevalonate decarboxylase | |
| 2625960 | -2.144 | citrate (Si)-synthase activivity, transferase activity, transferring acyl groups, acyl groups converted into alkyl on transfer | | Citrate synthase-like | |
| 2597019 | -2.177 | structural constituent of ribosome, acyl-CoA thioesterase activity | acyl-CoA metabolism, protein biosynthesis | Ribosomal protein, zinc-binding; Acyl-CoA thioesterase | |
| 2744685 | -2.191 | hydrolase activity, acting on carbon-nitrogen (but not peptide) bonds | nitrogen metabolism | Nitrilase/cyanide hydratase and apolipoprotein N-acyltransferase | |
| 2295738 | -2.194 | metal ion transporter activity, zinc ion transporter activity | zinc ion transport, metal ion transport | Zinc/iron permease | |
| 2459294 | -2.217 | glycine hydroxymethyltransferase activity | glycine metabolism, L-serine metabolism | Glycine hydroxymethyltransferase | |
| 2131899 | -2.260 | ATP binding | anion transport | Anion-transporting ATPase | |
| 67134 | -2.268 | catalytic activity, amylo-alpha-1,6-glucosidase activity | glycogen biosynthesis | Six-hairpin glycosidase-like; Amylo-alpha-1,6-glucosidase | |
| 2688579 | -2.281 | | | | |
| 2197094 | -2.283 | catalytic activity | metabolism | AMP-dependent synthetase and ligase | |
| 2742382 | -2.302 | copper ion binding, oxidoreductase activity | | Multicopper oxidase, type 2; Multicopper oxidase, type 3; | |

| | | | | | |
|---|---|---|---|---|---|
| | | | | Multicopper oxidase, copper-binding site | |
| 2676001 | -2.325 | hydrolase activity, hydrolyzing O-glycosyl compounds | carbohydrate metabolism | Glycoside hydrolase family 2, immunoglobulin-like beta-sandwich | |
| 2701053 | -2.359 | | | | |
| 2681988 | -2.366 | catalytic activity, malate dehydrogenase activity, binding, L-malate dehydrogenase activity, oxidoreductase activity | carbohydrate metabolism, TCA intermediate metabolism, malate metabolism, metabolism | Lactate dehydrogenase/glycoside hydrolase, family 4; Malate dehydrogenase, active site | |
| 2603451 | -2.369 | FAD binding, oxidoreductase activity | | Pyridine nucleotide-disulphide oxidoreductase, NAD-binding region | |
| 48253 | -2.383 | gamma-glutamyltransferase activity | | Gamma-glutamyltranspeptidase | |
| 2602715 | -2.384 | catalytic activity, monooxygenase activity, homoserine dehydrogenase activity, binding, iron ion binding, heme binding | electron transport, amino acid biosynthesis, metabolism | Cytochrome P450; Homoserine dehydrogenase, catalytic | |
| 2679415 | -2.395 | beta-galactosidase activity | carbohydrate metabolism | Glycoside hydrolase, family 35 | |
| 2257329 | -2.448 | oxidoreductase activity | potassium ion transport | Aldo/keto reductase; Potassium channel, voltage-dependent, beta subunit, KCNAB-related | |
| 2202445 | -2.466 | hydrogen-transporting ATPase activity, rotational mechanism, hydrogen ion transporter activity, hydrogen-transporting ATP synthase activity, rotational mechanism, hydrogen-exporting ATPase , activity, phosphorylative mechanism | energy coupled proton transport, against the electrochemical , ATP synthesis coupled proton transport | ATPase, F1/V1/A1 complex, alpha/beta subunit, nucleotide-binding | |
| 2663344 | -2.509 | protein binding, GTP binding | cell cycle | Septin; Cell division/GTP binding protein | Metabolism |
| 2677767 | -2.532 | nicotinate-nucleotide diphosphorylase (carboxylating) activity | nicotinamide adenine dinucleotide biosynthesis; pyridine nucleotide biosynthesis | Quinolinate phosphoribosyl transferase; Nicotinate-nucleotide pyrophosphorylase | |
| 2598507 | -2.539 | | | | |
| 2674496 | -2.541 | catalytic activity, pyrroline-5-carboxylate reductase activity, binding | electron transport, proline biosynthesis, metabolism | | |
| 2597536 | -2.544 | hydrogen-transporting ATPase activity, rotational mechanism | | ATPase, V0/A0 complex, subunit C/D | |
| 2598358 | -2.546 | catalytic activity, phosphoribosylaminoimidazolecarboxamide formyltransferase activity, IMP cyclohydrolase activity | IMP biosynthesis, purine nucleotide biosynthesis | AICARFT/IMPCHase bienzyme | |

| | | | | | |
|---|---|---|---|---|---|
| 2562064 | -2.568 | phosphoglycerate kinase activity | glycolysis | Phosphoglycerate kinase | |
| 2599726 | -2.644 | catalytic activity, FAD binding, binding, oxidoreductase activity | metabolism | Pyridine nucleotide-disulphide oxidoreductase, NAD-binding region | |
| 2489815 | -2.648 | catalytic activity, fumarate hydratase activity | fumarate metabolism | Fumarate lyase; L-Aspartase-like | |
| 2601378 | -2.653 | metalloexopeptidase activity, hydrolase activity | proteolysis and peptidolysis | Creatinase; Peptidase M24, catalytic core | |
| 2603431 | -2.656 | oxidoreductase activity | metabolism | Aldehyde dehydrogenase, conserved site; Aldehyde/histidinol dehydrogenase | |
| 2599489 | -2.668 | catalytic activity, binding, coenzyme binding, DNA binding | metabolism | NAD-dependent epimerase/dehydratase | |
| 2312113 | -2.757 | catalytic activity, binding, coenzyme binding | metabolism | NAD-dependent epimerase/dehydratase | |
| 2678377 | -2.782 | catalytic activity | metabolism | Transketolase | |
| 2582001 | -2.787 | hydrolase activity | | Hydantoinase/oxoprolinase | |
| 2163958 | -2.788 | | | | |
| 2745544 | -2.790 | NAD-dependent histone deacetylase activity, ATP binding, zinc ion binding, DNA binding, nucleoside triphosphatase activity, nucleotide binding, ATPase activity | regulation of transcription, DNA-dependent, protein amino acid deacetylation, chromatin silencing | NAD-dependent histone deacetylase, silent information regulator Sir2; ABC transporter-like; AAA+ ATPase, core | |
| 2599948 | -2.815 | catalytic activity, binding, ATP binding, oxidoreductase activity | fructose 2,6-bisphosphate metabolism, metabolism | Fructose-2,6-bisphosphatase; Short-chain dehydrogenase/reductase SD; Glucose/ribitol dehydrogenase | |
| 2501050 | -2.829 | 1-pyrroline-5-carboxylate dehydrogenase activity, oxidoreductase activity | proline biosynthesis, metabolism | Delta-1-pyrroline-5-carboxylate dehydrogenase 1; Aldehyde dehydrogenase | |
| 2750313 | -2.856 | hydrolase activity | | Nucleoside phosphatase GDA1/CD39 | |
| 2561893 | -2.871 | catalytic activity, transketolase activity | metabolism | Transketolase | |
| 2254211 | -2.914 | glutamate-ammonia ligase activity | glutamine biosynthesis, nitrogen metabolism | Glutamine synthetase, beta-Grasp | |
| 2599605 | -2.917 | metalloexopeptidase activity, oxidoreductase activity, acting on the CH-CH group of donors, catalytic activity, cofactor binding, methionyl aminopeptidase activity, binding, transporter activity, transition, metal ion binding, heme binding, acyl-CoA dehydrogenase activity | electron transport, proteolysis and peptidolysis, transport, metabolism | Peptidase M24, catalytic core; Acyl-CoA oxidase/dehydrogenase, type 1; AMP-dependent synthetase and ligase; Lipocalin; Cytochrome b5 | |
| 2676205 | -2.938 | transporter activity, oxidoreductase activity, acting on the aldehyde or oxo group of donors, disulfide as acceptor,oxoglutarate dehydrogenase (lipoamide) activity | glycolysis, transport, metabolism | Major facilitator superfamily; Dehydrogenase, E1 component; 2-oxoglutarate dehydrogenase, E1 component | |

| | | | | | |
|---|---|---|---|---|---|
| 2455051 | -2.949 | catalytic activity | metabolism | Thiolase-like | |
| 2597083 | -2.960 | acyltransferase activity | metabolism | Phospholipid/glycerol acyltransferase | |
| 2164570 | -3.018 | gamma-glutamyltransferase activity | | Gamma-glutamyltranspeptidase | |
| 2595545 | -3.029 | methionine adenosyltransferase activity, ATP binding | one-carbon compound metabolism | S-adenosylmethionine synthetase | |
| 2540747 | -3.046 | catalytic activity, FAD binding, oxidoreductase activity | | Berberine/berberine-like; FAD-binding, type 2 | |
| 2676916 | -3.103 | binding, 3,4 dihydroxy-2-butanone-4-phosphate synthase activity | vitamin B2 biosynthesis, transport | Mitochondrial substrate carrier; 3,4-Dihydroxy-2-butanone 4-phosphate synthase | |
| 2676596 | -3.125 | catalytic activity, carboxyl- and carbamoyltransferase activity, monooxygenase activity, binding, amino acid binding, oxidoreductase activity | electron transport, amino acid metabolism, aromatic compound metabolism, metabolism | Aspartate/ornithine carbamoyltransferase; Monooxygenase, FAD-binding; Glucose/ribitol dehydrogenase; Pyridine nucleotide-disulphide oxidoreductase, class I; Aromatic-ring hydroxylase; Short-chain dehydrogenase/reductase SDR | |
| 2146082 | -3.185 | oxidoreductase activity, acting on the CH-OH group of donors, NAD or NADP as acceptor, isocitrate dehydrogenase (NAD) activity | metabolism, tricarboxylic acid cycle | Isocitrate/isopropylmalate dehydrogenase | |
| 2675623 | -3.192 | ubiquitin-dependent protein catabolism | | Cullin | |
| 2685606 | -3.195 | ubiquitin-dependent protein catabolism | | Cullin | |
| 2092683 | -3.219 | catalytic activity | metabolism, trehalose biosynthesis | Glycosyl transferase, family 20; Trehalose-phosphatase; HAD-superfamily hydrolase, subfamily IIB | |
| 2679519 | -3.222 | catalytic activity, dihydroxy-acid dehydratase activity | branched chain family amino acid biosynthesis, metabolism | Dihydroxy-acid and 6-phosphogluconate dehydratase | |
| 2538519 | -3.244 | 5-methyltetrahydropteroyltriglutamate-homocysteine S-methyltransferase activity, zinc ion binding | methionine biosynthesis, amino acid biosynthesis | Methionine synthase, vitamin-B12 independent; Cobalamin (vitamin B12)-independent methionine synthase MetE, N-terminal; 5-methyltetrahydropteroyltriglutamate--homocysteine S-methyltransferase | |
| 2070913 | -3.269 | catalytic activity | carbohydrate metabolism | Glycosyl hydrolase, family 13, catalytic region | |
| 2707128 | -3.308 | catalytic activity, DNA binding | DNA methylation, metabolism | Ribulose-phosphate binding barrel; C-5 cytosine-specific DNA methylase | |
| 2111470 | -3.316 | transferase activity, transferring acyl groups, acyl groups converted into alkyl on transfer | | Citrate synthase-like | |
| 2642095 | -3.323 | triose-phosphate isomerase activity | metabolism | Triosephosphate isomerase | Metabolism |
| 2681810 | -3.323 | hydrolase activity, oxidoreductase activity | | Metallophosphoesterase; Aldo-keto reductase | |
| 2575823 | -3.346 | catalytic activity, oxidoreductase activity | electron transport, metabolism | Transketolase, C-terminal/Pyruvate-ferredoxin oxidoreductase, domain II; Flavoprotein pyridine nucleotide cytochrome reductase; | |

| | | | | | |
|---|---|---|---|---|---|
| | | | | Oxidoreductase FAD/NAD(P)-binding; FAD-binding, type 1; Pyruvate ferredoxin/flavodoxin oxidoreductase | |
| 2273629 | -3.357 | protein binding | cell communication | Phox-like | |
| 2592709 | -3.365 | DNA binding, nucleoside triphosphatase activity, nucleotide binding | DNA replication | DNA polymerase III clamp loader subunit; AAA+ ATPase, core | |
| 2624619 | -3.372 | D-amino-acid oxidase activity, oxidoreductase activity | | D-amino acid oxidase, conserved site; FAD dependent oxidoreductase | |
| 2676329 | -3.438 | catalytic activity, phospho-N-acetylmuramoyl-pentapeptide-transferase activity, isocitrate lyase activity | carboxylic acid metabolism, metabolism | Isocitrate lyase and phosphorylmutase; Pyruvate/Phosphoenolpyruvate kinase, catalytic core; Glycosyl transferase, family 4 | |
| 2683261 | -3.444 | electron-transferring-flavoprotein dehydrogenase activity, oxidoreductase activity | electron transport | Electron transfer flavoprotein-ubiquinone oxidoreductase; Alpha-helical ferredoxin | |
| 2602397 | -3.452 | catalytic activity, malate dehydrogenase activity, binding, oxidoreductase activity, acting on the CH-OH group of donors, NAD or NADP as , L-malate dehydrogenase activity, oxidoreductase activity | glycolysis, carbohydrate metabolism, TCA intermediate metabolism, malate metabolism, metabolism | Lactate dehydrogenase/glycoside hydrolase, family 4; Malate dehydrogenase, active site | |
| 2695856 | -3.459 | | | | |
| 2057820 | -3.514 | chitinase activity, hydrolase activity, hydrolyzing O-glycosyl compounds | chitin catabolism, carbohydrate metabolism | Chitinase II; Glycoside hydrolase, family 18, catalytic domain | |
| 2276847 | -3.517 | oxidoreductase activity | electron transport | NADH dehydrogenase (ubiquinone), 24 kDa subunit | |
| 2599260 | -3.539 | oxidoreductase activity, acting on the CH-OH group of donors, NAD or NADP as acceptor, 3-isopropylmalate dehydrogenase activity | metabolism, leucine biosynthesis | Isocitrate/isopropylmalate dehydrogenase; 3-isopropylmalate dehydrogenase | |
| 2592701 | -3.552 | catalytic activity, oxidoreductase activity, acting on CH-OH group of donors, binding, oxidoreductase activity, acting on the CH-OH group of donors, NAD or ADP as acceptor glycerol-3-phosphate dehydrogenase (NAD) activity oxidoreductase activity | glycerol-3-phosphate catabolism, glycerol-3-phosphate metabolism, carbohydrate metabolism, metabolism | NAD-dependent glycerol-3-phosphate dehydrogenase | |
| 2489166 | -3.626 | protein binding, GTP binding | cell cycle | Septin; Cell division/GTP binding protein | |
| 2560766 | -3.630 | binding | transport | Mitochondrial substrate carrier | |
| 2502569 | -3.662 | glycerophosphodiester phosphodiesterase activity | glycerol metabolism | Glycerophosphoryl diester phosphodiesterase | |
| 2676071 | -3.726 | catalytic activity, ATP binding, glutathione synthase activity | glutathione biosynthesis | PreATP-grasp-like fold, Glutathione synthase, substrate-binding, eukaryotic, Glutathione synthase, eukaryotic; PreATP-grasp-like fold | |

| | | | | | |
|---|---|---|---|---|---|
| 2750499 | -3.738 | catalytic activity, pyridoxal phosphate binding | cysteine biosynthesis from serine, metabolism | Pyridoxal phosphate-dependent enzyme, beta subunit; Cysteine synthase/cystathionine beta-synthase P-phosphate-binding site | |
| 2063627 | -3.761 | oxidoreductase activity | | FAD dependent oxidoreductase | |
| 2681708 | -3.793 | catalytic activity, 3-phosphoshikimate 1-carboxyvinyltransferase activity, hydroxymethylglutaryl-CoA reductase (NADPH) activity, shikimate 5-dehydrogenase activity, shikimate kinase activity, 3-dehydroquinate dehydratase activity, transferase activity, transferring alkyl or aryl (other than methyl) groups, 3-dehydroquinate synthase activity, binding, coenzyme binding, ATP binding | coenzyme A metabolism, aromatic amino acid family biosynthesis, metabolism | RNA 3'-terminal phosphate cyclase/enolpyruvate transferase, alpha/beta; 3-phosphoshikimate 1-carboxyvinyltransferase, subgroup; Hydroxymethylglutaryl-CoA reductase, class I/II, catalytic; Quinate/shikimate 5-dehydrogenase/glutamyl-tRNA reductase; Shikimate-5-dehydrogenase | |
| 2615440 | -3.798 | catalytic activity, binding, coenzyme binding | metabolism | NAD-dependent epimerase/dehydratase | |
| 2748159 | -3.802 | oxidoreductase activity, acting on the CH-CH group of donors, transition metal ion binding, heme binding, acyl-CoA dehydrogenase activity | electron transport, metabolism | Acyl-CoA dehydrogenase/oxidase; Cythocrome b5 | |
| 2700111 | -3.808 | catalytic activity | metabolism | Transketolase | |
| 2751395 | -3.829 | catalytic activity, succinate-CoA ligase (ADP-forming) activity, binding, zinc ion binding, ATP citrate synthase activity, transferase activity, transferring acyl groups, acyl groups converted into alkyl on transfer | metabolism | ATP-citrate lyase/succinyl-CoA ligase | |
| 2544123 | -3.858 | catalytic activity, GDP-mannose 4,6-dehydratase activity, binding, coenzyme binding, oxidoreductase activity | GDP-mannose metabolism, metabolism | NAD-dependent epimerase/dehydratase; GDP-mannose 4,6-dehydratase; Short-chain dehydrogenase/reductase SDR | |
| 2056817 | -3.862 | oxidoreductase activity | metabolism | Aldehyde dehydrogenase | |
| 2596037 | -3.869 | hydrogen-transporting ATPase activity, rotational mechanism, hydrogen ion transporter activity, nucleoside triphosphatase activity, nucleotide binding, hydrogen-transporting ATP, synthase activity, rotational mechanism, hydrogen-exporting ATPase activity, phosphorylative mechanism | ATP synthesis coupled proton transport, ATP biosynthesis | ATPase, F1/V1/A1 complex; AAA+ ATPase | |
| 2669280 | -3.872 | transporter activity, binding | transport | Adenine nucleotide translocator 1; Mitochondrial carrier protein | |
| 2675304 | -3.875 | beta-galactosidase activity | carbohydrate metabolism | Glycoside hydrolase, family 35 | |

| | | | | | |
|---|---|---|---|---|---|
| 2674424 | -3.897 | catalytic activity, binding, oxidoreductase activity | metabolism | Short-chain dehydrogenase/reductase SDR; Glucose/ribitol dehydrogenase | |
| 2675904 | -3.958 | hydrolase activity, hydrolyzing O-glycosyl compounds, oxidoreductase activity | carbohydrate metabolism | Glycoside hydrolase family 2, immunoglobulin-like beta-sandwich; Aldo/keto reductase | |
| 2595683 | -3.959 | | L-serine biosynthesis, metabolism | Phosphoserine aminotransferase; Aminotransferase, class V/Cysteine desulphurase | |
| 2596034 | -3.971 | | regulation of signal transduction, response to biotic stimulus | TAP42-like protein | |
| 2679035 | -3.975 | protein binding | | BAR | |
| 2611835 | -3.978 | catalytic activity | metabolism | Thiolase-like | |
| 2530747 | -4.039 | | | | |
| 2551610 | -4.084 | peroxidase activity, ATP binding, nucleoside triphosphatase activity, nucleotide binding, heme binding, ATPase activity, ATPase activity, coupled to transmembrane movement of substances | electron transport, transport, response to oxidative stress | ABC transporter, transmembrane region; AAA+ ATPase; ABC transporter-like | |
| 2469166 | -4.118 | | | | |
| 2600574 | -4.149 | nucleobase transporter activity, glycogen (starch) synthase activity | glycogen biosynthesis, nucleobase, nucleoside, nucleotide and nucleic acid transport | Permease for cytosine/purines, uracil, thiamine, allantoin; Glycogen synthase | |
| 1132839 | -4.163 | oxidoreductase activity | metabolism | Aldehyde dehydrogenase; Aldehyde dehydrogenase, conserved site | |
| 2567541 | -4.163 | catalytic activity, isocitrate lyase activity | carboxylic acid metabolism, metabolism | Isocitrate lyase and phosphorylmutase; Pyruvate/Phosphoenolpyruvate kinase, catalytic core | |
| 2274136 | -4.182 | | | | |
| 2695493 | -4.213 | oxidoreductase activity | carbohydrate metabolism, metabolism | Aldehyde dehydrogenase; Carbohydrate kinase, FGGY | |
| 2583245 | -4.218 | serine carboxypeptidase activity, serine-type peptidase activity | proteolysis and peptidolysis | Peptidase S10, serine carboxypeptidase; Peptidase S26A, signal peptidase I | |
| 2603678 | -4.254 | nucleoside triphosphatase activity, nucleotide binding, ATPase activity, ATP binding | | AAA+ ATPase, core; ABC transporter-like | |
| 2599939 | -4.292 | catalytic activity, magnesium ion binding | | Thiamine pyrophosphate enzyme, C-terminal TPP-binding | |
| 2595597 | -4.347 | protein kinase activity, protein-tyrosine kinase activity, protein serine/threonine kinase activity, ATP binding | protein amino acid phosphorylation | Protein kinase; Serine/threonine protein kinase-related; Tyrosine protein kinase; Serine/threonine protein kinase | |
| 2728614 | -4.348 | hydrogen-transporting ATPase activity, rotational mechanism, hydrogen-transporting ATP synthase activity, rotational mechanism | ATP synthesis coupled proton transport, proton-transporting two-sector | ATPase, F0 complex, subunit B, mitochondrial | |

| | | | | | |
|---|---|---|---|---|---|
| | | | ATPase complex | | |
| 2680688 | -4.351 | phosphorylase activity, pyridoxal phosphate binding | carbohydrate metabolism | Glycosyl transferase, family 35; Glycogen/starch/alpha-glucan phosphorylase | |
| 2158355 | -4.395 | | | | |
| 2697037 | -4.400 | catalytic activity, cofactor binding, oxidoreductase activity, acting on the CH-OH group of donors, NAD or NADP as acceptor, binding | metabolism | NAD(P)-binding; D-isomer specific 2-hydroxyacid dehydrogenase, NAD-binding; D-isomer specific 2-hydroxyacid dehydrogenase, catalytic region; | |
| 2271828 | -4.451 | catalytic activity, binding, oxidoreductase activity, zinc ion binding | metabolism | NAD(P)-binding; Alcohol dehydrogenase, zinc-containing, conserved site | |
| 2530104 | -4.453 | oxidoreductase activity | | NADH:flavin oxidoreductase/NADH oxidase | |
| 2681688 | -4.466 | transferase activity, transferring nitrogenous groups, pyridoxal phosphate binding | biosynthesis | Aminotransferase, class I and II | |
| 2546713 | -4.535 | catalytic activity, binding | metabolism | NAD(P)-binding | Metabolism |
| 2666754 | -4.609 | glutamate-ammonia ligase activity | glutamine biosynthesis, nitrogen metabolism | Glutamine synthetase, beta-Grasp | |
| 2282069 | -4.668 | oxidoreductase activity | metabolism | MaoC-like dehydratase | |
| 2473140 | -4.673 | catalytic activity, calcium ion binding, alpha-amylase activity | carbohydrate metabolism | Glycosyl hydrolase, family 13, subfamily, catalytic region; Alpha-amylase, fungi | |
| 2703628 | -4.678 | monooxygenase activity, heme binding, iron ion binding | electron transport | Cytochrome P450 | Metabolism |
| 2677337 | -4.679 | catalase activity | electron transport, response to oxidative stress | Catalase | |
| 2470775 | -4.686 | serine carboxypeptidase activity | proteolysis and peptidolysis | Peptidase S10, serine carboxypeptidase | |
| 2596885 | -4.724 | methylmalonate-semialdehyde dehydrogenase (acylating) activity, oxidoreductase activity | valine metabolism, metabolism | Methylmalonate-semialdehyde dehydrogenase; Aldehyde dehydrogenase | |
| 2274225 | -4.775 | metal ion binding, superoxide dismutase activity | superoxide metabolism | Manganese and iron superoxide dismutase | |
| 2662238 | -4.794 | adenylosuccinate synthase activity, GTP binding | purine nucleotide biosynthesis | Adenylosuccinate synthetase | |
| 2645499 | -4.800 | | | | |
| 2680115 | -4.821 | catalytic activity | | UBA/THIF-type NAD/FAD binding fold | |
| 2695732 | -4.848 | transferase activity, transferring nitrogenous groups, pyridoxal phosphate binding | biosynthesis | Aminotransferase, class I and II | |
| 2003623 | -4.914 | ribose-phosphate diphosphokinase activity, magnesium ion binding, transferase activity | ribonucleoside monophosphate biosynthesis, nucleotide biosynthesis, nucleoside metabolism | Phosphoribosyl pyrophosphokinase | |
| 2591856 | -4.926 | transporter activity | transport | Major facilitator superfamily | |

| | | | | | |
|---|---|---|---|---|---|
| 2547111 | -4.995 | thiamin-phosphate diphosphorylase activity, hydroxyethylthiazole kinase activity | thiamin biosynthesis | Thiamine monophosphate synthase, Hydroxyethylthiazole kinase | |
| 2598771 | -4.999 | oxidoreductase activity | electron transport | FMN-dependent alpha-hydroxy acid dehydrogenase; FMN-dependent alpha-hydroxy acid dehydrogenase, active site | |
| 2611769 | -5.029 | heme binding, iron ion binding | electron transport, tricarboxylic acid cycle | CybS | |
| 2535945 | -5.049 | phosphoribosylaminoimidazolesuccinocarboxamide synthase activity | purine nucleotide biosynthesis | SAICAR synthetase | |
| 2707033 | -5.109 | oxidoreductase activity | | Aldo/keto reductase | |
| 2598986 | -5.182 | catalytic activity, binding, oxidoreductase activity, zinc ion binding | regulation of transcription, DNA-dependent, metabolism | Alcohol dehydrogenase, zinc-containing, conserved site | |
| 2255769 | -5.204 | | | Translationally controlled tumour-associated TCTP | |
| 2116057 | -5.247 | acid phosphatase activity | | Histidine acid phosphatase | |
| 2674598 | -5.262 | dihydrolipoamide S-succinyltransferase activity; acyltransferase activity | metabolism, tricarboxylic acid cycle | Dihydrolipoamide succinyltransferase; Catalytic domain of components of various dehydrogenase complexes | |
| 2546053 | -5.277 | oxidoreductase activity | | Aldo/keto reductase | |
| 2569226 | -5.329 | oxidoreductase activity | | Aldo/keto reductase | |
| 2600373 | -5.429 | oxidoreductase activity, acting on sulfur group of donors, disulfide as acceptor, transcription factor activity | regulation of transcription, DNA-dependent, protein metabolism | Methionine sulphoxide reductase A; Homeobox | |
| 2750135 | -5.558 | glucosamine-6-phosphate deaminase activity | N-acetylglucosamine metabolism, carbohydrate metabolism | Glucosamine-6-phosphate isomerase | |
| 2511932 | -5.613 | catalytic activity, binding, oxidoreductase activity | electron transport, metabolism | Oxidoreductase | |
| 2745344 | -5.690 | protein binding, zinc ion binding, electron carrier activity | electron transport | Zinc finger, RING-type; Zinc finger, variant RING-type; Electron transfer flavoprotein, beta-subunit, core | |
| 2704408 | -5.718 | glucose-6-phosphate isomerase activity | glycolysis, gluconeogenesis | Phosphoglucose isomerase (PGI) | |
| 2679247 | -5.747 | catalytic activity, binding, oxidoreductase activity | fatty acid metabolism, metabolism | 3-hydroxyacyl-CoA dehydrogenase, NAD binding; 6-phosphogluconate dehydrogenase | |
| 2508662 | -5.825 | cysteine-type endopeptidase activity | proteolysis and peptidolysis | Peptidase C1B, bleomycin hydrolase | |
| 2562773 | -5.905 | beta-N-acetylhexosaminidase activity | carbohydrate metabolism | Glycoside hydrolase, family 20 | |
| 2679037 | -5.936 | GTP binding, oxidoreductase activity | cell cycle | Cell division/GTP binding protein; Redoxin | |
| 2026034 | -6.087 | protein kinase activity, protein-tyrosine kinase activity, protein serine/threonine kinase activity, ATP binding | protein amino acid phosphorylation | Protein kinase; Serine/threonine protein kinase-related; Protein kinase ATP binding, conserved site | |

| | | | | | |
|---|---|---|---|---|---|
| 2623304 | -6.190 | chitinase activity, hydrolase activity, hydrolyzing O-glycosyl compounds | chitin catabolism, carbohydrate metabolism | Chitinase II; Glycoside hydrolase, family 18, catalytic domain | |
| 2602081 | -6.224 | catalytic activity, binding | metabolism | NAD(P)-binding | |
| 2512669 | -6.257 | catalytic activity, adenosylhomocysteinase activity, binding | one-carbon compound metabolism, metabolism | S-adenosyl-L-homocysteine hydrolase | |
| 2744912 | -6.324 | hydrolase activity, hydrolyzing O-glycosyl compounds | carbohydrate metabolism | Glycoside hydrolase, family 3, N-terminal; Glycoside hydrolase, family 3, C-terminal | |
| 2706632 | -6.417 | carbohydrate binding, chitinase activity, hydrolase activity, hydrolyzing O-glycosyl compounds | chitin catabolism, carbohydrate metabolism | Carbohydrate-binding family V/XII; Chitinase II; Glycoside hydrolase, family 18, catalytic domain | |
| 2549499 | -6.520 | catalytic activity, alpha,alpha-trehalose-phosphate synthase (UDP-forming) activity | trehalose biosynthesis | Glycosyl transferase, family 20; Alpha,alpha-trehalose-phosphate synthase | |
| 2491930 | -6.956 | | | | |
| 2703762 | -6.978 | protein kinase activity, protein serine/threonine kinase activity, ATP binding | actin filament polymerization, protein amino acid phosphorylation | Protein kinase, core; Serine/threonine protein kinase; Serine/threonine protein kinase; ARP23 complex 20 kDa subunit; Protein kinase, core | |
| 2338336 | -7.163 | catalytic activity | metabolism | Isocitrate lyase and phosphorylmutase; Pyruvate/Phosphoenolpyruvate kinase, catalytic core | |
| 2637783 | -7.899 | catalytic activity, carboxy-lyase activity, magnesium ion binding | | Thiamine pyrophosphate enzyme, central region; Pyruvate decarboxylase/indolepyruvate decarboxylase | Metabolism |
| 2679467 | -3.696 | metalloexopeptidase activity, hydrolase activity | vitamin B2 biosynthesis, transport | Peptidase M24, catalytic core, Creatinase | |
| 2599143 | -6.084 | catalytic activity | metabolism | Thiolase-like | |
| 2753281 | -2.004 | metalloexopeptidase activity | proteolysis and peptidolysis | Peptidase M24, catalytic core | |
| 2601059 | -2.109 | catalytic activity, nucleic acid binding, ATP binding, acyltransferase activity | metabolism | Carbamoyl phosphate synthetase, large subunit, ATP-binding; Polynucleotidyl transferase, Ribonuclease H fold; Acyltransferase ChoActase/COT/CPT | |
| 2598891 | -2.219 | | | | |
| 2596927 | -2.226 | nucleic acid binding, ATP binding, helicase activity, sugar porter activity, transporter activity, ATP dependent helicase activity | carbohydrate transport, transport | DNA/RNA helicase;General substrate transporter; Major facilitator superfamily; Sugar transporter, conserved site | |
| 2532844 | -2.249 | | | | |
| 2679553 | -2.254 | N-acetyltransferase activity | metabolism | GCN5-related N-acetyltransferase | |
| 2597780 | -2.374 | catalytic activity, sugar porter activity, protein serine/threonine phosphatase activity, transporter activity | carbohydrate transport, protein amino acid dephosphorylation, transport | Protein phosphatase 2C-related; Major facilitator superfamily; Sugar transporter, conserved site | |
| 2600743 | -2.451 | protein binding, oxidoreductase activity | | BTB/POZ fold; Aldo/keto reductase | |

| | | | | | |
|---|---|---|---|---|---|
| 2597208 | -2.456 | GTP binding, phosphopyruvate hydratase activity | glycolysis | GTP1/OBG; GTP-binding protein, HSR1-related; Enolase | |
| 1166793 | -2.478 | structural constituent of ribosome | protein biosynthesis | Ribosomal protein L32e | |
| 2055515 | -2.689 | | | | |
| 2678297 | -2.697 | GTP binding | | GTP-binding protein, HSR1-related; Conserved hypothetical protein CHP00092 | |
| 2596562 | -2.801 | catalytic activity, binding, phosphogluconate dehydrogenase (decarboxylating) activity, sugar porter activity, transporter activity, oxidoreductase activity | carbohydrate transport, transport, pentose-phosphate shunt, metabolism | 6-phosphogluconate dehydrogenase, C-terminal-like; Sugar transporter; General substrate transporter; Major facilitator superfamily | |
| 2596102 | -2.854 | catalytic activity, structural constituent of ribosome, magnesium ion binding, phospholipid-translocating ATPase activity, ATP binding, ATPase activity, coupled to transmembrane movement of ions, phosphorylative mechanism | phospholipid transport, protein biosynthesis, transport, metabolism | Haloacid dehalogenase-like hydrolase; Ribosomal protein S2; Phospholipid-translocating P-type ATPase, flippase | |
| 2676133 | -2.998 | FAD binding, oxidoreductase activity, acting on CH-OH group of donors | | FAD binding;oxidoreductase activity, acting on CH-OH group of donors | |
| 2681776 | -3.056 | aminopeptidase activity | proteolysis and peptidolysis | Peptidase M17, leucyl aminopeptidase | |
| 2677686 | -3.223 | catalytic activity, transferase activity, transferring nitrogenous groups, GTPase activity, GTP binding, structural constituent of nuclear pore, pyridoxal phosphate binding | biosynthesis | Aminotransferase, class I and II; Dynamin GTPase effector; Nucleoporin, Nsp1-like, C-terminal | |
| 2605570 | -3.271 | oxidoreductase activity | | Aldo/keto reductase | |
| 2746421 | -3.367 | GTP binding | protein transport, small GTPase mediated signal transduction | Ran GTPase; Ras GTPase; Ras small GTPase, Ras type; Ras small GTPase, Rho type; Ras small GTPase, Rab type; Small GTP-binding protein; | |
| 2501262 | -3.380 | lipid metabolism, metabolism | | Patatin; Acyl transferase/acyl hydrolase/lysophospholipase | |
| 2254126 | -3.407 | | | | |
| 2565304 | -3.574 | catalytic activity | | Esterase/lipase/thioesterase | |
| 2676814 | -3.683 | | metabolism | Aminotransferase, class V/Cysteine desulphurase | |
| 2693747 | -3.683 | transporter activity, binding | signal transduction, transport | Rho GTPase activation protein; Lipocalin | |
| 2608284 | -3.721 | carboxypeptidase A activity, chaperone activator activity, ATPase stimulator activity, zinc ion binding | proteolysis and peptidolysis | Peptidase M14; Activator of Hsp90 ATPase | |
| 2643023 | -3.727 | FAD binding, oxidoreductase activity, acting on CH-OH group of donors | | Glucose-methanol-choline oxidoreductase | |

| | | | | | |
|---|---|---|---|---|---|
| 2601683 | -3.841 | catalytic activity | metabolism | Haloacid dehalogenase-like hydrolase | |
| 2590711 | -4.009 | oxidoreductase activity | | Aldo/keto reductase | |
| 2242729 | -4.078 | | | | |
| 2674209 | -4.126 | GTPase activity, GTP binding | | Dynamin, GTPase region | |
| 2502834 | -4.190 | hydrolase activity | | Dienelactone hydrolase | |
| 2573839 | -4.196 | GTPase activity, GTP binding | | Dynamin, GTPase region | |
| 2674809 | -4.359 | phosphoric ester hydrolase activity, nucleic acid binding, zinc ion binding | carbohydrate metabolism | Inositol phosphatase/fructose-1,6-bisphosphatase | |
| 2750138 | -4.398 | FAD binding, oxidoreductase activity, acting on CH-OH group of donors | alcohol metabolism | Glucose-methanol-choline oxidoreductase | |
| 2595836 | -4.624 | catalytic activity, binding, oxidoreductase activity | metabolism | Short-chain dehydrogenase/reductase SDR; Glucose/ribitol dehydrogenase | |
| 2198127 | -4.702 | | | | |
| 2674907 | -4.808 | catalytic activity, GTP binding, magnesium ion binding, phospholipid-translocating ATPase activity, ATP binding, ATPase activity, coupled to transmembrane movement of ions, phosphorylative mechanism | cell cycle, phospholipid transport, transport, metabolism | Cell division/GTP binding protein; Phospholipid-translocating P-type ATPase, flippase; ATPase, P-type, K/Mg/Cd/Cu/Zn/Na/Ca/Na/H-transporter; Phospholipid-translocating P-type ATPase, flippase | |
| 2604988 | -4.877 | endocytosis | | Adaptin ear-binding coat-associated protein 1 NECAP-1 | |
| 2548824 | -4.927 | FAD binding, oxidoreductase activity, acting on CH-OH group of donors | alcohol metabolism | Glucose-methanol-choline oxidoreductase | |
| 2677972 | -5.017 | structural constituent of ribosome, GTP binding | protein transport, small GTPase mediated signal transduction, protein biosynthesis | Ribosomal protein S7e; Ras small GTPase, Ras type; Ras small GTPase, Rho type; Ras small GTPase, Rab type; Small GTP-binding protein | |
| 2596033 | -5.487 | FAD binding, oxidoreductase activity, acting on CH-OH group of donors, GTP binding | protein transport, small GTPase mediated signal transduction | Glucose-methanol-choline oxidoreductase; Ras GTPase; Ras small GTPase, Ras type; Ras small GTPase, Rab type | |
| 2600127 | -5.602 | | | | |
| 2485740 | -5.725 | | | | |
| 2669543 | -5.767 | GTP binding | protein transport, small GTPase mediated signal transduction | Ras small GTPase, Ras type; Ras small GTPase, Rho type; Ras small GTPase, Rab type; Small GTP-binding protein | |
| 2599947 | -5.771 | | | | |
| 2676541 | -5.918 | | | | |
| 2676989 | -5.951 | cysteine protease inhibitor activity | | Proteinase inhibitor I25, cystatin | |
| 2675760 | -6.184 | hydrolase activity | | Dienelactone hydrolase | |
| 2696064 | -6.313 | FAD binding, oxidoreductase activity, acting on CH-OH group of donors | alcohol metabolism | Glucose-methanol-choline oxidoreductase | |

| | | | | | |
|---|---|---|---|---|---|
| 2688236 | -6.589 | catalytic activity, binding, ATP binding, nucleotide binding, tRNA ligase activity, oxidoreductase activity | amino acid activation, protein biosynthesis, metabolism | Aminoacyl-tRNA synthetase, class I, conserved site; Short-chain dehydrogenase/reductase SDR; Glucose/ribitol dehydrogenase | |
| 2615761 | -6.668 | | thiamin biosynthesis | Thiamine biosynthesis Thi4 protein | |
| 2660812 | -7.757 | oxidoreductase activity | negative regulation of transcription | Flavodoxin/nitric oxide synthase | |
| 2573733 | -7.983 | FAD binding, oxidoreductase activity, acting on CH-OH group of donors | alcohol metabolism | Glucose-methanol-choline oxidoreductase | |
| 2576488 | -2.017 | | | | |
| 2485364 | -2.026 | | | | |
| 2611978 | -2.044 | aminopeptidase activity, prolyl aminopeptidase activity | proteolysis and peptidolysis | Peptidase S33, prolyl aminopeptidase | |
| 2695299 | -2.050 | nucleobase, nucleoside, nucleotide kinase activity, nucleotide kinase activity, ATP binding, phosphotransferase activity, phosphate group as acceptor | | | |
| 2528625 | -2.056 | | | | |
| 2597024 | -2.073 | catalytic activity, ATP binding | metabolism | ATP-citrate lyase/succinyl-CoA ligase, Succinyl-CoA synthetase, beta subunit, ATP-grasp fold | |
| 2675881 | -2.073 | protein binding, GTP binding, nucleic acid binding, GTPase activity | nucleocytoplasmic transport, signal transduction, small GTPase mediated signal transduction, protein transport, intracellular protein transport | Ras small GTPase, Ras type; Ras small GTPase, Rho type; Ras small GTPase, Rab type; Ran GTPase; Small GTP-binding protein | |
| 2593151 | -2.085 | | | | |
| 2674968 | -2.087 | nucleic acid binding, ATP binding, zinc ion binding | | Zinc finger, C2H2-type; Dephospho-CoA kinase | |
| 2662384 | -2.137 | hydrolase activity, hydrolyzing O-glycosyl compounds | carbohydrate metabolism | Glycoside hydrolase, family 7 | |
| 2679354 | -2.251 | catalytic activity, carbamoyl-phosphate synthase activity, ATP binding | metabolism, nitrogen metabolism | | |
| 2510342 | -2.269 | protein binding | cell communication | Phox-like | |
| 2602331 | -2.285 | | | | |
| 2747169 | -2.302 | catalytic activity, aspartate-semialdehyde dehydrogenase activity, binding, zinc ion binding, NADPH binding, protein dimerization activity, oxidoreductase activity, acting on the aldehyde or oxo group of donors,, NAD or NADP as acceptor, transcription factor activity | amino acid metabolism, methionine biosynthesis, amino acid biosynthesis, threonine biosynthesis, metabolism, regulation of transcription, DNA-dependent | NAD(P)-binding; Aspartate-semialdehyde dehydrogenase, conserved site | |

| | | | | | |
|---|---|---|---|---|---|
| 2674282 | -2.317 | catalytic activity, binding, NAD(P) transhydrogenase activity, NAD(P) transhydrogenase (AB-specific) activity, oxidoreductase activity | electron transport, proton transport, metabolism | NAD(P) transhydrogenase, beta subunit; Alanine dehydrogenase/PNT | |
| 2563170 | -2.346 | | | | |
| 2565612 | -2.407 | | | | |
| 2597428 | -2.437 | catalytic activity, binding | metabolism | NAD(P)-binding | |
| 2597932 | -2.443 | | | | |
| 2675450 | -2.572 | catalytic activity, hydrolase activity, acting on carbon-nitrogen (but not peptide) bonds | nucleosome assembly, carbohydrate metabolism | Glycoside hydrolase/deacetylase, beta/alpha-barrel; Polysaccharide deacetylase; Nucleosome assembly protein (NAP) | |
| 2675749 | -2.623 | | | | |
| 2598403 | -2.664 | catalytic activity | | Six-hairpin glycosidase-like | |
| 2681415 | -2.699 | spore wall assembly (sensu Bacteria), pathogenesis | | Delta-endotoxin CytB | |
| 2599051 | -2.714 | nucleic acid binding, integrase activity | mitochondrial genome maintenance | Integrase, C-terminal, retroviral; Mitochondrial genome maintenance MGM101 | |
| 2679667 | -2.736 | acid phosphatase activity, nucleic acid binding, zinc ion binding | | Histidine; Zinc finger, C2H2-type | |
| 2601385 | -2.817 | binding | transport | Mitochondrial substrate carrier | |
| 2586510 | -2.843 | hydrolase activity, acting on carbon-nitrogen (but not peptide) bonds | | Metal-dependent hydrolase, composite | |
| 2700276 | -2.859 | subtilase activity, serine-type peptidase activity | proteolysis and peptidolysis | Peptidase S8 and S53, subtilisin, kexin, sedolisin | |
| 2599659 | -2.880 | phosphoenolpyruvate carboxykinase (ATP) activity, phosphoenolpyruvate carboxykinase activity, ATP binding, purine nucleotide binding | gluconeogenesis | Phosphoenolpyruvate carboxykinase, ATP-utilising | |
| 2677212 | -2.958 | catalytic activity | metabolism | Haloacid dehalogenase-like hydrolase | |
| 2701600 | -3.050 | cysteine-type endopeptidase activity, nutrient reservoir activity | proteolysis and peptidolysis | Peptidase, cysteine peptidase active site; Cupin 1 | |
| 2469527 | -3.052 | | | | |
| 2699394 | -3.060 | | | | |
| 2597062 | -3.081 | mannose-1-phosphate guanylyltransferase (GDP) activity, nucleotidyltransferase activity | polysaccharide metabolism, biosynthesis | Mannose-6-phosphate isomerase, type II; Nucleotidyl transferase | |
| 2677111 | -3.102 | | | | |
| 2696330 | -3.170 | catalytic activity, FAD binding, protein-tyrosine kinase activity, ATP binding, oxidoreductase activity | protein amino acid phosphorylation | FAD-linked oxidase, Tyrosine protein kinase | |
| 1141646 | -3.183 | GTPase activity, structural molecule activity, GTP binding | microtubule-based movement, microtubule-based process | Alpha tubulin | |
| 2676241 | -3.187 | catalytic activity, transcriptional repressor activity, binding | regulation of nitrogen | NmrA-like | |

| | | | | | |
|---|---|---|---|---|---|
| | | | utilization, metabolism | | |
| 2473950 | -3.195 | | | | |
| 2555894 | -3.261 | | | | |
| 2579864 | -3.265 | | | | |
| 1157787 | -3.270 | nucleotidyltransferase activity | metabolism | UTP--glucose-1-phosphate uridylyltransferase | |
| 2608058 | -3.272 | binding | | Armadillo-type fold | |
| 2677861 | -3.282 | catalytic activity | | Six-hairpin glycosidase-like | |
| 2542644 | -3.297 | | | | |
| 2580701 | -3.303 | two-component sensor molecule activity, protein-histidine kinase activity, two-component response regulator activity, signal transducer activity, ATP binding, transferase activity, transferring phosphorus-containing groups | signal transduction, phosphorylation, regulation of transcription, DNA-dependent, two-component signal transduction system (phosphorelay), peptidyl-histidine phosphorylation | Signal transduction histidine kinase | |
| 2546333 | -3.321 | protein binding | | BTB/POZ-like | |
| 2476052 | -3.360 | | | | |
| 2528310 | -3.393 | | | | |
| 2597310 | -3.410 | protein binding, peptidyl-prolyl cis-trans isomerase activity | protein folding | Peptidyl-prolyl cis-trans isomerase, cyclophilin-type; BTB/POZ-like | |
| 2673553 | -3.442 | catalytic activity, hydrolase activity, acting on carbon-nitrogen (but not peptide) bonds | carbohydrate metabolism | Polysaccharide deacetylase; Glycoside hydrolase/deacetylase, beta/alpha-barrel | |
| 2679088 | -3.462 | hydrolase activity, hydrolyzing O-glycosyl compounds | carbohydrate metabolism | Glycoside hydrolase, family 16 | |
| 2032525 | -3.463 | binding | transport | Mitochondrial substrate carrier | |
| 2450251 | -3.466 | binding | transport | Mitochondrial substrate carrier | |
| 2081064 | -3.474 | phosphoric monoester hydrolase activity | metabolism | HAD-superfamily hydrolase, subfamily IB, PSPase-like | |
| 2596805 | -3.495 | motor activity, binding, ATP binding | | Myosin head, motor region | |
| 2580789 | -3.495 | | | | |
| 2666791 | -3.497 | serine-type peptidase activity | proteolysis and peptidolysis | Peptidase S28 | |
| 2600933 | -3.529 | beta-1,4-mannosylglycoprotein 4-beta-N-acetylglucosaminyltransferase activity | N-linked glycosylation | Glycosyl transferase, family 17 | |
| 2596101 | -3.551 | oxidoreductase activity, acting on the CH-CH group of donors, zinc ion binding,transcription factor activity, acyl-CoA dehydrogenase activity | electron transport, regulation of transcription, DNA-dependent, metabolism | Acyl-CoA dehydrogenase/oxidase; Fungal transcriptional regulatory protein | |
| 2742326 | -3.566 | | | | |
| 2567746 | -3.642 | | | | |
| 2743762 | -3.643 | racemase and epimerase activity, acting on amino acids and derivatives | metabolism | Asp/Glu racemase | |

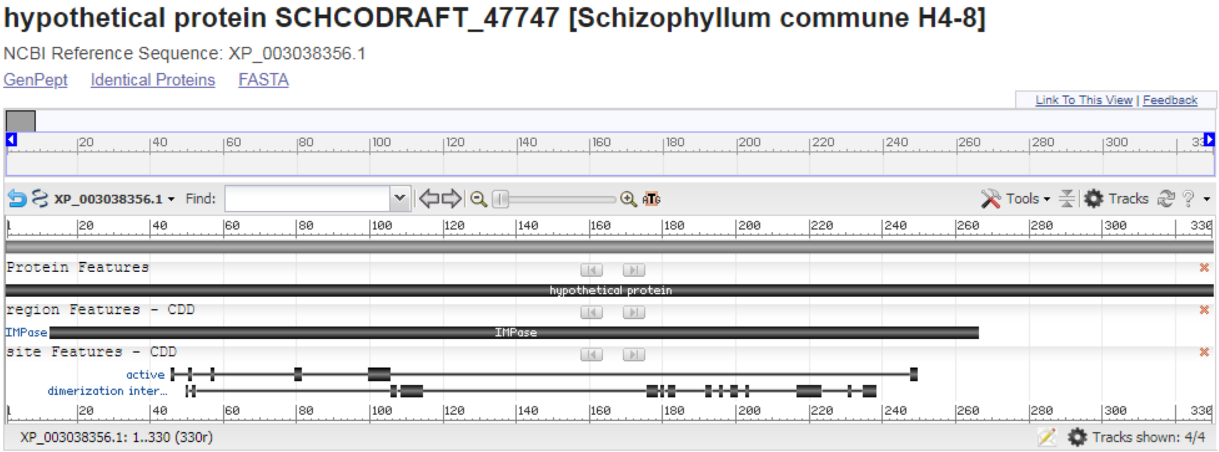
| | | | | | |
|---|---|---|---|---|---|
| 1126744 | -3.656 | | | | |
| 2513636 | -3.699 | | | | |
| 2480617 | -3.702 | transcription factor activity, signal transducer activity | signal transduction, regulation of transcription, DNA-dependent | STAT transcription factor, coiled coil | |
| 1186194 | -3.731 | | | | |
| 2629150 | -3.755 | | | | |
| 2678193 | -3.778 | monooxygenase activity, oxidoreductase activity | electron transport, aromatic compound metabolism, metabolism | Monooxygenase, FAD-binding; Aromatic-ring hydroxylase | |
| 2597110 | -3.784 | | | | |
| 2678233 | -3.789 | catalytic activity, binding, oxidoreductase activity, ketol-acid reductoisomerase activity | branched chain family amino acid biosynthesis, metabolism | 6-phosphogluconate dehydrogenase; Acetohydroxy acid isomeroreductase | |
| 2703645 | -3.807 | | | | |
| 2676356 | -3.808 | catalytic activity, thiosulfate sulfurtransferase activity, chorismate mutase activity, carbon-carbon lyase activity | cell redox homeostasis, aromatic compound metabolism, sulfate transport, aromatic amino acid family biosynthesis | Pyruvate/Phosphoenolpyruvate kinase, catalytic core; Thiosulphate sulphurtransferase, conserved site; Chorismate mutase | |
| 2512880 | -3.882 | | | | |
| 2599179 | -3.888 | subtilase activity | serine-type peptidase activity | Peptidase S8 and S53, subtilisin, kexin, sedolisin; Peptidase S53, propeptide | |
| 2595924 | -4.020 | | | | |
| 2517922 | -4.026 | binding | | Armadillo-type fold | |
| 2675392 | -4.052 | catalytic activity, oxidoreductase activity, acting on CH-OH group of donors, nucleic acid binding, binding, zinc ion binding, oxidoreductase activity, acting on the CH-OH group of donors, NAD or NADP as acceptor, glycerol-3-phosphate dehydrogenase (NAD) activityoxidoreductase activity | glycerol-3-phosphate catabolism, glycerol-3-phosphate metabolism, carbohydrate metabolism, metabolism | NAD(P)-binding; NAD-dependent glycerol-3-phosphate dehydrogenase, C-terminal; NAD-dependent glycerol-3-phosphate dehydrogenase, N-terminal; NAD-dependent glycerol-3-phosphate dehydrogenase; 6-phosphogluconate dehydrogenase, C-terminal-like; Zinc finger, CCCH-type | |
| 2705026 | -4.086 | catalytic activity, glyceraldehyde-3-phosphate dehydrogenase (phosphorylating) activity, binding, glyceraldehyde-3-phosphate dehydrogenase activity | glycolysis, glucose metabolism, metabolism | Glyceraldehyde-3-phosphate dehydrogenase, type I; NAD(P)-binding | |
| 2708788 | -4.089 | protein kinase activity, protein-tyrosine kinase activity, protein serine/threonine kinase activity, ATP binding | protein amino acid phosphorylation | Protein kinase; Serine/threonine protein kinase-related; Tyrosine protein kinase | |

| | | | | | |
|---|---|---|---|---|---|
| 2585457 | -4.110 | peroxidase activity, heme binding | electron transport, response to oxidative stress | Haem peroxidase | |
| 2702757 | -4.120 | protein binding | | BTB/POZ fold | |
| 2675838 | -4.137 | asparagine synthase (glutamine-hydrolyzing) activity, nucleic acid binding | asparagine biosynthesis | Asparagine synthase; RNA recognition, region 1; Prohibitin | |
| 2689658 | -4.163 | | | | |
| 2508404 | -4.184 | | | | |
| 2536636 | -4.186 | | | | |
| 2676276 | -4.193 | 3-deoxy-7-phosphoheptulonate synthase activity, binding | biosynthesis, transport, aromatic amino acid family biosynthesis | DHAP synthase, class 1; Mitochondrial substrate carrier | |
| 2601164 | -4.212 | structural constituent of ribosome, FAD binding, oxidoreductase activity, ureidoglycolate hydrolase activity, protein binding, ATP binding, DNA binding, allantoicase activity | protein biosynthesis, electron transport, regulation of transcription, DNA-dependent, intra-Golgi transport, allantoin catabolism | Ribosomal protein S9; Pyridine nucleotide-disulphide oxidoreductase, NAD-binding region; Ureidoglycolate hydrolase; SMCs flexible hinge | |
| 2636005 | -4.251 | | | | |
| 2601836 | -4.269 | protein binding | | Leucine-rich repeat | |
| 2747968 | -4.283 | | | | |
| 2677721 | -4.357 | nucleic acid binding, zinc ion binding, DNA binding, transcription factor activity, pyridoxal phosphate binding | amino acid metabolism, regulation of transcription, DNA-dependent, transcription | RNA recognition motif, RNP-1; Fungal transcriptional regulatory protein; Cys/Met metabolism, pyridoxal phosphate-dependent enzyme | |
| 2600547 | -4.450 | catalytic activity, carboxyl- and carbamoyltransferase activity, ATP binding, aspartate carbamoyltransferase activity, carbamoyl-phosphate synthase, amino acid binding | glutamine metabolism, amino acid metabolism, biosynthesis, metabolism, 'de novo' pyrimidine base biosynthesis, nitrogen metabolism | Glutamine amidotransferase superfamily; Anthranilate synthase component II/delta crystallin; Carbamoyl phosphate synthase, GATase region | |
| 2500167 | -4.479 | nucleoside triphosphatase activity, nucleotide binding, ATPase activity, ATP binding | | AAA+ ATPase; ABC transporter-like | |
| 2589858 | -4.479 | hydrolase activity, hydrolyzing O-glycosyl compounds | carbohydrate metabolism | Glycoside hydrolase, family 6 | |
| 2668578 | -4.498 | metallopeptidase activity, protein dimerization activity, hydrolase activity | proteolysis and peptidolysis | Peptidase M20; Peptidase M20, dimerisation | |
| 2740159 | -4.555 | carbohydrate binding, hydrolase activity, hydrolyzing O-glycosyl compounds | carbohydrate metabolism | Carbohydrate-binding family V/XII | |

| | | | | | |
|---|---|---|---|---|---|
| 2619084 | -4.564 | | | | |
| 2675792 | -4.572 | catalytic activity, histone deacetylase activity | histone deacetylation | Histone deacetylase | |
| 2750360 | -4.579 | | | | |
| 2600930 | -4.611 | catalytic activity, binding, oxidoreductase activity | metabolism | Short-chain dehydrogenase/reductase SDR; Glucose/ribitol dehydrogenase; Aromatic-ring hydroxylase | |
| 2530078 | -4.612 | aminopeptidase activity, prolyl aminopeptidase activity | proteolysis and peptidolysis | Peptidase S33, prolyl aminopeptidase | |
| 2702164 | -4.676 | cysteine-type endopeptidase activity, manganese ion binding, pyrophosphatase activity, hydrolase activity | proteolysis and peptidolysis | Peptidase, cysteine peptidase active site; Phosphoesterase, RecJ-like; DHHA2 | |
| 2599114 | -4.683 | rRNA processing | | Small-subunit processome, Utp13 | |
| 2336543 | -4.723 | | | Legume-like lectin | |
| 2195418 | -4.728 | | | | |
| 2191162 | -4.729 | aminopeptidase activity, prolyl aminopeptidase activity | proteolysis and peptidolysis | Peptidase S33, prolyl aminopeptidase | |
| 2538728 | -4.745 | ribonuclease activity, RNA binding | | Ribonuclease II and R | |
| 2692813 | -4.756 | catalytic activity | metabolism | Mandelate racemase/muconate lactonizing enzyme | |
| 1138359 | -4.807 | | | | |
| 2600484 | -4.855 | | | | |
| 2574569 | -4.905 | | | | |
| 2540154 | -4.920 | | | | |
| 2526241 | -4.921 | binding | | Armadillo-type fold | |
| 2677843 | -5.051 | prenyltransferase activity, nucleic acid binding, Rho GDP-dissociation inhibitor activity, zinc ion binding | | RHO protein GDP dissociation inhibitor; UbiA prenyltransferase | |
| 2675318 | -5.056 | | | | |
| 2680457 | -5.116 | zinc ion binding | | Zinc finger, MYND-type | |
| 2596872 | -5.152 | | | | |
| 2539960 | -5.166 | hydrolase activity, hydrolyzing O-glycosyl compounds | carbohydrate metabolism | Glycoside hydrolase, family 3 | |
| 2603513 | -5.244 | catalytic activity, structural constituent of ribosome, binding, transporter activity | protein biosynthesis, transport, metabolism | Ribosomal protein L1; Major intrinsic protein | |
| 2184213 | -5.272 | | | | |
| 2283081 | -5.275 | | metabolism | Aminotransferase, class V/Cysteine desulphurase | |
| 2456797 | -5.306 | 1-aminocyclopropane-1-carboxylate deaminase activity, catalytic activity, pyridoxal phosphate binding | metabolism, amine catabolism | 1-aminocyclopropane-1-carboxylate deaminase; Pyridoxal phosphate-dependent enzyme, beta subunit | |
| 2117151 | -5.337 | | | | |
| 2753424 | -5.357 | | | | |
| 2599803 | -5.375 | glutamate decarboxylase activity, carboxy-lyase activity, transcription factor activity, pyridoxal phosphate binding | carboxylic acid metabolism, glutamate metabolism, regulation of transcription, DNA-dependent | Pyridoxal phosphate-dependent decarboxylase; Glutamate decarboxylase; Homeobox | |

| | | | | | |
|---|---|---|---|---|---|
| 2592036 | -5.404 | acid phosphatase activity | | Histidine acid phosphatase | |
| 2738673 | -5.412 | GTP binding, | protein transport, small GTPase mediated signal transduction, | Ras GTPase; Ras small GTPase Ras type, Ras small GTPase, Rho type; Ras small GTPase, Rab type; Ras GTPase | |
| 2603182 | -5.417 | galactokinase activity, phosphotransferase activity, alcohol group as acceptor, ATP binding, kinase activity | carbohydrate phosphorylation, phosphorylation, metabolism, galactose metabolism, | Galactokinase; Mevalonate and galactokinase; Mevalonate and galactokinase; GHMP kinase; GHMP kinase, ATP-binding, conserved site | |
| 2667616 | -5.439 | nucleic acid binding, exonuclease activity | | Putative 5-3 exonuclease | |
| 2572813 | -5.440 | protein folding | | Heat shock protein DnaJ | |
| 2597177 | -5.453 | isomerase activity, protein binding, oxidoreductase activity | electron transport | Peptidyl-prolyl cis-trans isomerase, PpiC-type | |
| 2500013 | -5.488 | transition metal ion binding, motor activity, heme binding, ATP binding, transferase activity, transferring hexosyl groups | | Myosin head, motor region; Cythochrome b5 | |
| 2600338 | -5.499 | | | | |
| 2508802 | -5.543 | | | | |
| 2548110 | -5.629 | | | | |
| 2667521 | -5.672 | calcium ion binding, phosphatidylserine decarboxylase activity | phospholipid biosynthesis | Calcium-binding EF-hand; Phosphatidylserine decarboxylase | |
| 2586789 | -5.751 | | | | |
| 2596097 | -5.780 | acid phosphatase activity | | Histidine acid phosphatase | |
| 2103552 | -5.812 | signal transducer activity | | Regulator of G protein signalling | |
| 2601160 | -5.815 | | | | |
| 2353446 | -5.828 | hydrogen-transporting ATPase activity, rotational mechanism, hydrogen-transporting ATP synthase activity, rotational mechanism | ATP synthesis coupled proton transport | ATPase, F1 complex, OSCP/delta subunit | |
| 2734322 | -5.850 | | | | |
| 2598879 | -5.897 | catalytic activity, phosphoric monoester hydrolase activity, transcription factor activity, hydrolase activity, oxidoreductase activity | regulation of transcription, DNA-dependent metabolism | Haloacid dehalogenase-like hydrolase; 2-phosphoglycolate phosphatase, eukaryotic; Homeobox; Aldo/keto reductase | |
| 2301961 | -5.998 | voltage-gated potassium channel activity | potassium ion transport | KCNAB voltage-gated K+ channel, beta subunit | Metabolism |
| 2565384 | -6.041 | catalytic activity, binding, oxidoreductase activity | metabolism | 6-phosphogluconate dehydrogenase | |
| 2692586 | -6.098 | hydrolase activity, hydrolyzing O-glycosyl compounds | carbohydrate metabolism | Glycoside hydrolase, family 3 | |
| 2535816 | -6.323 | | | | |
| 2620107 | -6.363 | | | | |
| 2630944 | -6.372 | | | | |
| 2264463 | -6.380 | hydrolase activity, hydrolyzing O-glycosyl compounds | carbohydrate metabolism | Glycoside hydrolase, family 17 | |
| 2697931 | -6.460 | | | | |

| | | | | | |
|---|---|---|---|---|---|
| 2439795 | -6.590 | | | | |
| 2676857 | -6.629 | catalytic activity | | Six-hairpin glycosidase-like | |
| 2498164 | -6.687 | hydrolase activity | | Alpha/beta hydrolase | |
| 2672812 | -6.774 | | | | |
| 2594802 | -6.799 | | | | |
| 2601175 | -6.817 | zinc ion binding, pepsin A activity, DNA binding, aspartic-type endopeptidase activity, transcription factor activity | regulation of transcription, DNA-dependent, proteolysis and peptidolysis, transcription | Fungal specific transcription factor; Peptidase A1 | |
| 2537084 | -6.822 | | | | |
| 1209921 | -7.028 | | | | |
| 2678455 | -7.083 | | | | |
| 2600628 | -7.098 | | | | |
| 2595350 | -7.335 | GTP binding | protein transport, small GTPase mediated signal transduction | Ras small GTPase, Ras type; Ras small GTPase, Rho type; Ras small GTPase, Rab type; Small GTP-binding protein | |
| 2574999 | -7.973 | catalytic activity, protein kinase activity, ATP binding | protein amino acid phosphorylation | Six-hairpin glycosidase-like; Protein kinase ATP binding, conserved site | |
| 2595248 | -7.989 | serine carboxypeptidase activity | proteolysis and peptidolysis | Peptidase S10, serine carboxypeptidase | |
| 2452507 | -9.017 | | | | |
| 2640541 | -9.051 | | | | |



hypothetical protein SCHCODRAFT_47747 [Schizophyllum commune H4-8]

NCBI Reference Sequence: XP_003038356.1

Figure S1. A screen-shot from NCBI shows a hypothetical protein inositol monophosphatase consists of 330 AA along with predicted active site: 46, 51, 57, 80-81,100-105, 248-249 and predicted dimerization interface (polypeptide binding site): 50, 52,106-107,109-114,176-178,180,182-183,192-193,196,199-200, 203, 217- 223, 231, 235-238.

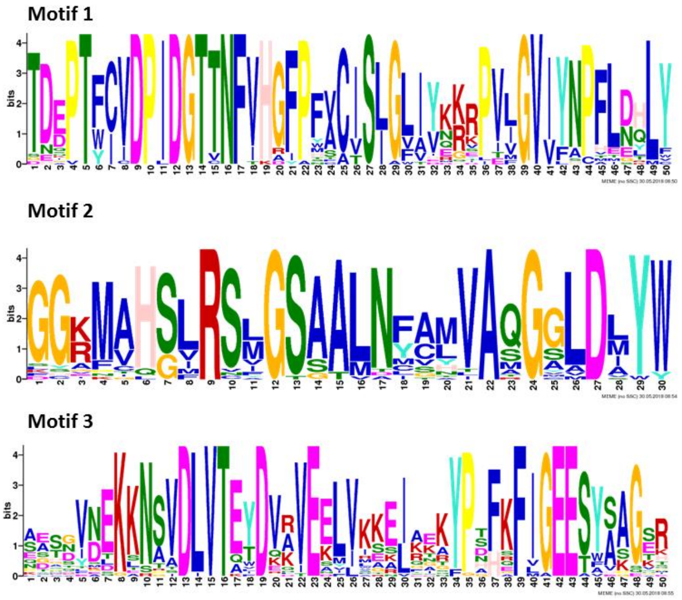
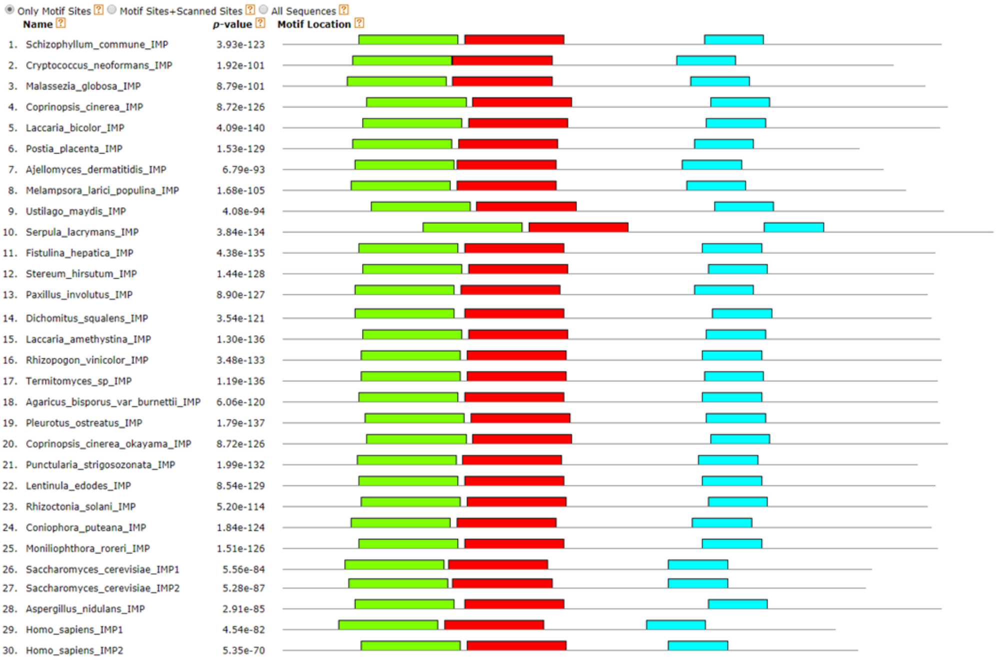


Figure S2. Three Motif search by MEME was performed using 31 IMP protein sequences from different organisms. These three motifs show a conserved superfamily, namely FIG (FBPase / IMPase / glpX-like domain). This superfamily represents a metal-dependent phosphatases enzyme with multivariant substrates. Fructose1,6-bisphospatase and the glpX-encoded variant is known to hydrolyze fructose 1,6-bisphosphate to fructose 6-phosphate. Meanwhile, inositol monophosphatases and the related enzyme group inositol polyphosphatases play crucial roles in eukaryotic signaling. These enzymes are sensitive to lithium and take part in the metabolism of the prominent messenger inositol 1,4,5-triphosphate ($IP_3$).


● Only Motif Sites ? ○ Motif Sites+Scanned Sites ? ○ All Sequences ?

| | Name ? | *p*-value ? |
|---|---|---|
| 1. | Schizophyllum_commune_IMP | 3.93e-123 |
| 2. | Cryptococcus_neoformans_IMP | 1.92e-101 |
| 3. | Malassezia_globosa_IMP | 8.79e-101 |
| 4. | Coprinopsis_cinerea_IMP | 8.72e-126 |
| 5. | Laccaria_bicolor_IMP | 4.09e-140 |
| 6. | Postia_placenta_IMP | 1.53e-129 |
| 7. | Ajellomyces_dermatitidis_IMP | 6.79e-93 |
| 8. | Melampsora_larici_populina_IMP | 1.68e-105 |
| 9. | Ustilago_maydis_IMP | 4.08e-94 |
| 10. | Serpula_lacrymans_IMP | 3.84e-134 |
| 11. | Fistulina_hepatica_IMP | 4.38e-135 |
| 12. | Stereum_hirsutum_IMP | 1.44e-128 |
| 13. | Paxillus_involutus_IMP | 8.90e-127 |
| 14. | Dichomitus_squalens_IMP | 3.54e-121 |
| 15. | Laccaria_amethystina_IMP | 1.30e-136 |
| 16. | Rhizopogon_vinicolor_IMP | 3.48e-133 |
| 17. | Termitomyces_sp_IMP | 1.19e-136 |
| 18. | Agaricus_bisporus_var_burnettii_IMP | 6.06e-120 |
| 19. | Pleurotus_ostreatus_IMP | 1.79e-137 |
| 20. | Coprinopsis_cinerea_okayama_IMP | 8.72e-126 |
| 21. | Punctularia_strigosozonata_IMP | 1.99e-132 |
| 22. | Lentinula_edodes_IMP | 8.54e-129 |
| 23. | Rhizoctonia_solani_IMP | 5.20e-114 |
| 24. | Coniophora_puteana_IMP | 1.84e-124 |
| 25. | Moniliophthora_roreri_IMP | 1.51e-126 |
| 26. | Saccharomyces_cerevisiae_IMP1 | 5.56e-84 |
| 27. | Saccharomyces_cerevisiae_IMP2 | 5.28e-87 |
| 28. | Aspergillus_nidulans_IMP | 2.91e-85 |
| 29. | Homo_sapiens_IMP1 | 4.54e-82 |
| 30. | Homo_sapiens_IMP2 | 5.35e-70 |

Figure S3. The block diagram shows motif matches on 31 input sequence from different organisms. The length of the line (depicted in colored boxes green, red, and cyan) in different motifs show the length of a sequence relative to all the other sequences. Significant motifs were searched automatically using MEME (http://meme-suite.org/tools/meme) and the similarity was confirmed by BLAST (https://blast.ncbi.nlm.nih.gov/Blast.cgi?PAGE=Proteins). Green blocks indicate the 3rd motif, red blocks show the 1st motif and cyan block depicts the 2nd motif.

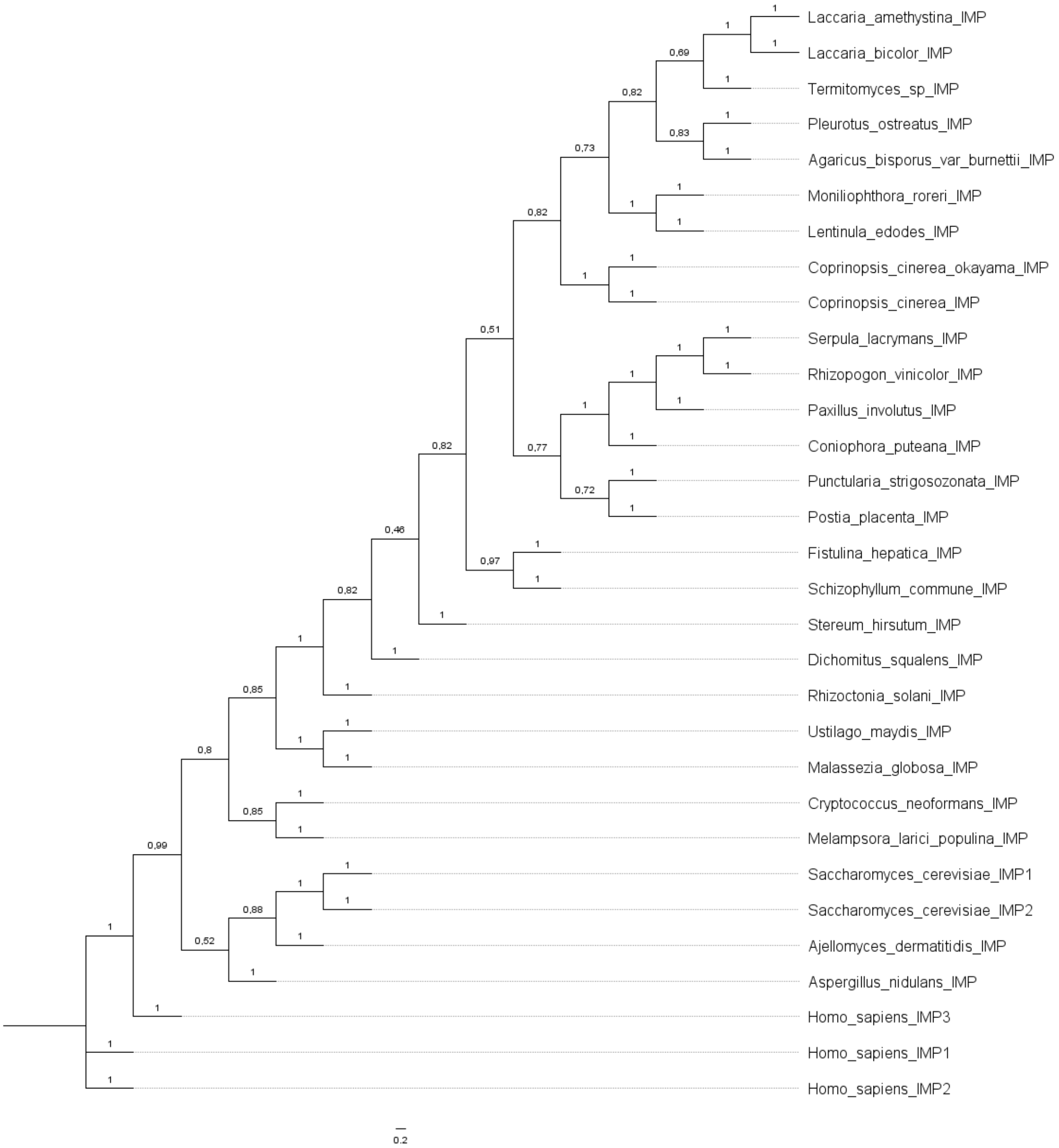


Figure S4. Phylogeny based on coding sequence information of inositol monophosphatase. Tree calculation was performed with Mr. Bayes via online CIPRUS Science Gateway after the alignment file was converted to NEXUS format.

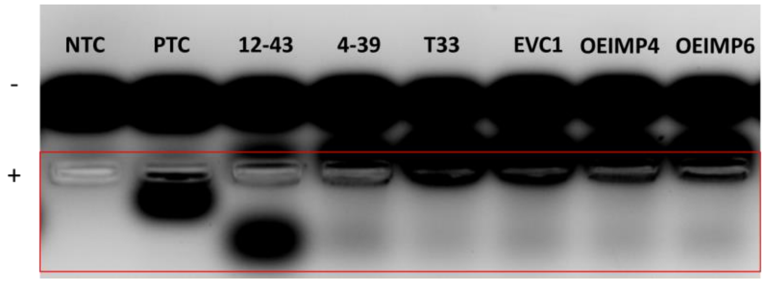


Figure S5. Protein kinase C activity of *imp* overexpressing mutants. Phosphorylated and unphosphorylated substrate peptides was separated by gel electrophoresis. Phosphorylated substrate peptide contains negative charge and thus. it runs to the anode (bottom), on the other hand the unphosphorylated substrate peptide contains positive charge, therefore it runs to the cathode (up). The gel showed in this picture is a representative gel. The assay was repeated two times to confirm the result. NTC is negative control and PTC is positive control.

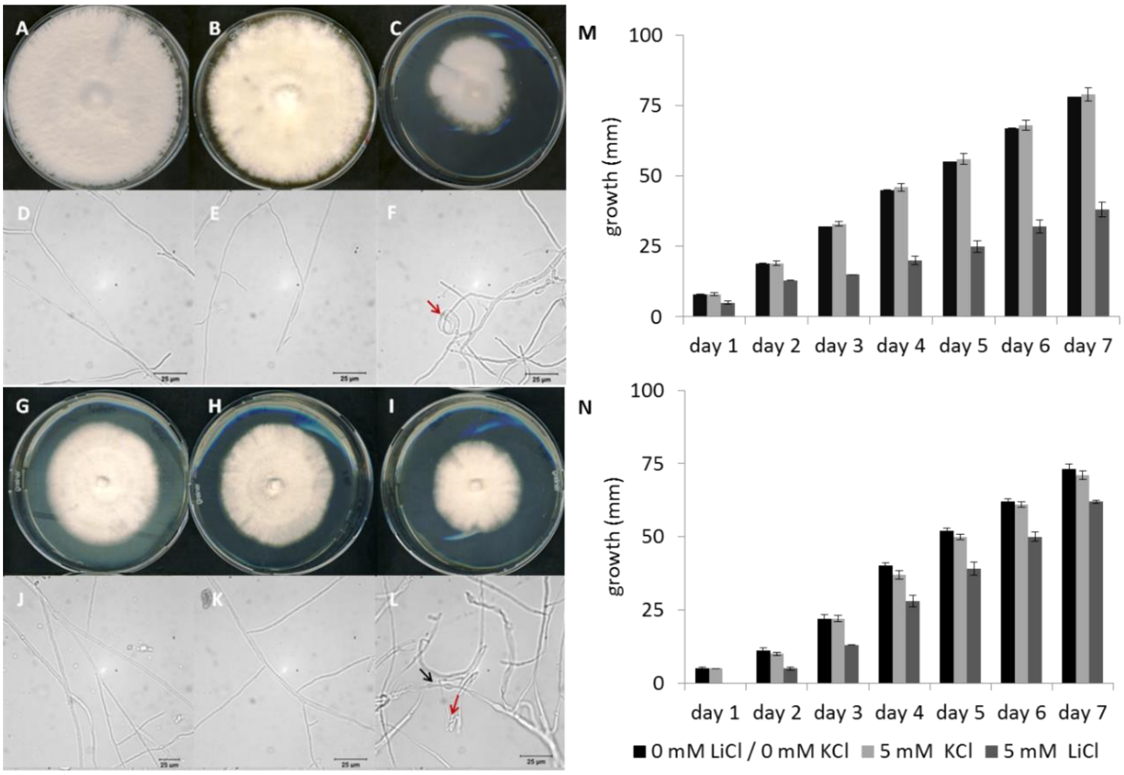


Figure S6. Morphology of *S. commune* wild type 4-39 is shown in A-C and constitutively active Ras1 mutant *ras1*$^{G12V}$ is depicted in G-I after 7-9 days cultivation at 30°C on CYM plate. Control without additional 5 mM LiCl and 5 mM KCl (A, G). control with 5 mM KCl (B, H). and treatment of 5 mM LiCl (C, I). Hyphal morphology of wild type 4-39 (D-F) and *ras1*$^{G12V}$ (J-L). Control without additional LiCl and KCl (D, J), control with 5 mM KCl (E, K). and treatment of 5 mM LiCl (F, L). Red arrow shows the folded-formed hyphae and black arrow shows swollen hyphae indicating that lithium is a stressor for the hyphae. infrequently multi-branching phenomenon was observed in the fungus treated with 5 mM LiCl. Fungal growth rates of *S. commune* wild type 4-39 (M) and constitutively active Ras1 mutant *ras1*$^{G12V}$ (N) in the presence of 5 mM KCl as salt control and treatment of 5 mM LiCl.

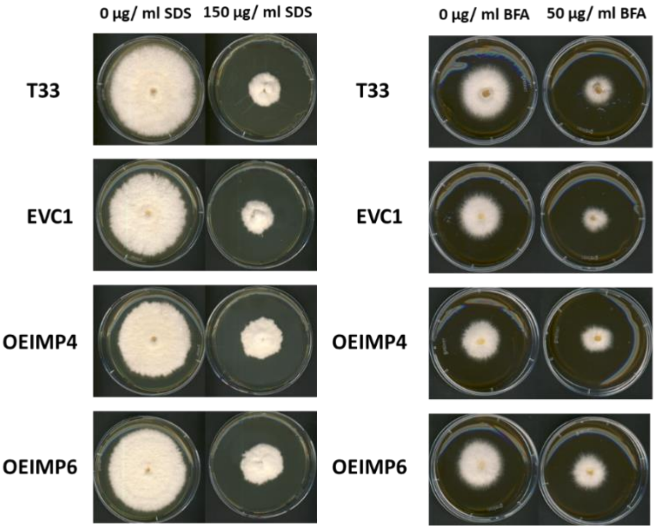


Figure S7. Morphology of overexpressing imp of *S. commune* prior to sodium dodecyl sulfate (SDS) effect and Brefeldin A (BFA) effect. SDS is a detergent that ruptures cell membrane and denature proteins in the cell membrane and BFA is an antibiotic that block vesicular transport along the secretory pathway.

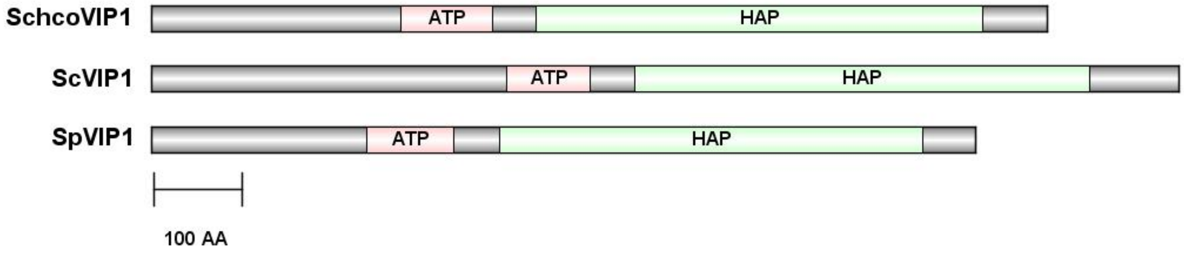


Figure S8. Domain organization of the SchcoVIP1 (VIP1 belongs to *S. commune*; schco3 2621615). Schematic alignment of the ATP grasp (ATP) and the histidine acid phosphatase (HAP) domains within the VIP proteins from *Saccharomyces cerevisiae* NP_013514 (ScVIP1). *Schizosaccharomyces pombe* NP_587877 (SpVIP1).

# 10 Abbreviations

| | |
|---|---|
| 1D | One dimension |
| 2D | Two dimensions |
| *act* | gene coding for actin |
| AGC | Automatic gain control |
| APS | Ammonium persulfate |
| BSA | Bovine Serum Albumin |
| CTAB | Cetyl trimethyl ammonium bromide |
| CW | Calcofluor-white |
| CYM | Complete Yeast Medium |
| CYM-T | Complete Yeast Medium supplemented with tryptophan |
| DAG | Diacylglycerol |
| DAPI | 4'-6-Diamidino-2-phenylindole |
| DMSO | Dimethyl sulfoxide |
| dpi | Day post inoculation |
| DNA | Deoxyribonucleic acid |
| DNAse | Deoxyribonuclease |
| DTT | Dithiothreitol |
| EDTA | Ethylenediaminetetraacetic acid |
| ER | Endoplasmic reticulum |
| FDR | False discovery rate |
| HCCA | Cyano-4-hydroxy-cinnamic acid |
| HEPES | 4-(2-hydroxyethyl)-1-piperazineethanesulfonic acid |
| HPLC | High liquid performance chromatography |
| MES | 2-(N-morpholino) ethanesulfonic acid |
| IMP | Inositol monophosphatase |
| *imp* | gene of inositol monophosphatase |
| Imk | Inositol phosphate multi kinase |
| Ipk | Inositol pentakishosphate 2-kinase |
| $IP_1$ | inositol monophosphate |
| $IP_2$ | inositol bisphosphate |
| $IP_3$ | inositol trisphosphate |
| $IP_4$ | inositol.tetrakisphosphate |
| $IP_5$ | inositol pentakisphosphate |
| $IP_6$ | inositol hexakisphosphate |
| $IP_7$ | diphosphoinositol pentakisphosphate |
| $IP_8$ | bis-diphosphoinositol tetrakisphosphate |
| ITmax | maximum injection time |

| | |
|---|---|
| LC | Liquid chromatography |
| LiCl | Lithium chloride |
| LoeD | *S. commune*, Loenen dominant monokaryon |
| MALDI TOF/MS | matrix-assisted laser desorption ionization times-of-flight mass spectrometry |
| MM | Minimal medium |
| MM-U | Minimal medium supplemented with uracil |
| MMU-T | Minimal medium supplemented with uracil and tryptophan |
| MS | Mass spectrometry |
| NCE | Normalized collision energy |
| PAGE | Polyacrylamide gel |
| PEG | Polyethylene glycol |
| PI | Phosphatidylinositol |
| Pi | Inorganic phosphate |
| PIP | Phosphatidylinositol phosphate |
| *pis* | gene phosphatidylinositol synthase |
| PCR | Polymerase chain reaction |
| PKC | protein kinase C |
| PLC | Phospholipase C |
| PIP2 | Phosphatidylinositol 4.5 bisphosphate |
| rcf | relative centrifugal force |
| RNA | Ribonucleic acid |
| RNAse | Ribonuclease |
| rpm | rotation per minute |
| RT | Room temperature |
| SAX-HPLC | Strong anion exchange - high liquid performance chromatography |
| SDS | Sodium dodecylsulfate |
| TatD | *S. commune,* Tattone dominant monokaryon |
| *tef* | gene coding for translation elongation factor EF1a |
| TAE | Tris acetate ethylenediaminetetraacetic acid |
| TBE | Tris borate ethylenediaminetetraacetic acid |
| TEMED | Tetramethylethylenediamine |
| TFA | Trifluoroacetic acid |
| *ubi* | gene coding for ubiquitin |
| UV | Ultraviolet |
| YMM | Yeast minimal media |
| YPD | Yeast extract peptone dextrose |

# Statement of authorship

This dissertation was done independently by me and nothing else than the mentioned references and sources were used.
I have not submitted this thesis as a test paper for any governmental or other form of scientific examination. Moreover, I have not tried to submit this thesis or an in major parts similar thesis as a dissertation to another university.

Diese Dissertation habe ich selbständig verfasst und keine anderen als die von mir angegebenen Referenz und Quellen benutzt.
Ich habe diese Dissertation noch nicht als Prüfungsarbeit für eine staatliche oder andere wissenschaftliche Prüfung eingereicht. Ferner habe ich nicht versucht diese Arbeit oder eine in wesentlichen Teilen ähnliche oder eine andere Abhandlung bei einer anderen Hochschule als Dissertation einzureichen.

Jena,12th of September 2018

…………………………………

Reyna Murry

# Curriculum vitae

## Personal information

| | |
|---|---|
| First name / Surname | Reyna Murry |
| Nationality | Indonesia |
| Place & Date of Birth | Bandung, 22nd October 1987 |
| Occupational Field | Microbiology |

## Work experiences

| | |
|---|---|
| Dates | May 2013 - May 2019 |
| Occupation or position held | Doctoral Researcher (PhD Student) |
| Main activities and responsibilities | Research on inositol phosphates signaling in *Schizophyllum commune* |
| Name of employer | Institute of Microbiology - Neugasse 25, 07743 Jena, Germany |

| | |
|---|---|
| Dates | June - July 2017, August - September 2017 |
| Occupation or position held | Doctoral Researcher (research stay) |
| Main activities and responsibilities | Research on inositol phosphates detection using SAX - HPLC |
| Name of employer | LMCB - MRC Laboratory for Molecular Biology,<br>University College London, UK |

| | |
|---|---|
| Dates | October 2016 - December 2016 |
| Occupation or position held | Intern |
| Main activities and responsibilities | Quality assurance for Click Chemistry Department |
| Name of employer | Jena Bioscience GmbH., Löbstedter str. 71, 07749 Jena, Germany |

| | |
|---|---|
| Dates | April 2016 - June 2016 |
| Occupation or position held | Intern |
| Main activities and responsibilities | Research and development in protein crystallography |
| Name of employer | Jena Bioscience GmbH., Löbstedter str. 71, 07749 Jena, Germany |

## Education and training

| | |
|---|---|
| Dates | October 2010 - March 2013 |
| Title of qualification awarded | Master Degree of Science |
| Principal subjects | Microbiology |
| Name of University | Friedrich Schiller University of Jena |

| | |
|---|---|
| Dates | September 2005 - January 2010 |
| Title of qualification awarded | Bachelor Degree of Science |
| Principal subjects | Biology |
| Name of University | Padjadjaran University, Bandung, Indonesia |

## Language skills

| | |
|---|---|
| Mother tongue | Bahasa Indonesia (Indonesian language) |
| Other language(s) | Malay (Near native)<br>English (Fluent)<br>German (Very good command) |

## Teaching skills and competences

Supervision of a student assistant (March 2017 - April 2018)

Supervision of a student assistant (August - October 2017)

Supervision of practical course Master Microbiology “Microbial interactions: Genetics and molecular biology in the higher basidiomycete *Schizophyllum commune*” (January 2017 - February 2017)

Supervision of a master student of Microbiology (July 2016 - April 2017)

Supervision of an apprentice (July - September 2015)

Supervision of practical course Master Microbiology “Microbial interactions: Genetics and molecular biology in the higher basidiomycete *Schizophyllum commune*” (October 2014 - February 2015)

Supervision of an apprentice (July - September 2014)

Supervision of practical course Master Microbiology “Microbial interactions: Genetics and molecular biology in the higher basidiomycete *Schizophyllum commune*” (October 2013 - February 2014)

## Organizational skills and competences

Organization team of Microbiology Conference (April 2014 - March 2015)

Coordinator of Indonesian Student Association Thuringia (February 2013 - March 2014)

Committee of Biology Student Association, Padjadjaran University (June 2005 - December 2009)

## Publication

Murry, R., Kniemeyer, O, Krause K, Saiardi, A, Kothe, E. 2019. Crosstalk between Ras and Inositol Phosphate signaling revealed by lithium action on inositol monophosphatase in *Schizophyllum commune.* Advances in Biological Regulation. Volume 72, May 2019, Pages 78-88.

## Conference and Symposium participation

### Talks

Murry, R., and Kothe, E. 2013. Crosstalk and Interrelation between Ras and phosphatidylinositol signaling via inositol monophosphatase in *Schizophyllum commune. International Leibniz Research School (ILRS) Seminar Group*, Jena, Germany.

Murry, R., and Kothe, E. 2013. Exploration of Inositol monophosphatase in

*Schizophyllum commune. 6th International Leibniz Research School (ILRS) Symposium*, Jena, Germany.

Murry, R., and Kothe, E. 2015. Crosstalk and interrelation between phosphatidylinositol and Ras signaling pathway via inositol monophosphatase of *Schizophyllum commune. 8th International Leibniz Research School (ILRS) Symposium*, Jena, Germany.

### Poster

Murry, R., Freihorst, D., and Kothe, E. 2013. Phosphatidylinositol (PI) signaling in the basidiomycete *Schizophyllum commune. 17th Signal Transduction Society (STS) Meeting*, Weimar, Germany.

Murry, R., and Kothe, E. 2014. Exploration of phosphatidylinositol signaling in the basidiomycete *Schizophyllum commune. 6th International Conference on Microbial Communication (MiCom)*, Jena, Germany.

Murry, R., and Kothe, E. 2014. Cross-talk between Ras- and PI- signaling through inositol monophosphatase in *Schizophyllum commune. 10th International Mycological Congress (IMC10)*, Bangkok, Thailand.

Murry, R., and Kothe, E. 2014. Cross-talk between Ras and Phosphatidylinositol (PI) signaling in *Schizophyllum commune. ILRS Symposium*, Jena, Germany.

Murry, R., and Kothe, E. 2015. Interrelation between Ras and Phosphatidylinositol signaling in the basidiomycete *Schizophyllum commune. The 28th Fungal Genetics Conference.* Pacific Grove, CA, USA.

Murry, R., and Kothe, E. 2015. Interrelation between Ras and Phosphatidylinositol signaling in the basidiomycete *Schizophyllum commune. 7th International Conference on Microbial Communication (MiCom)*, Jena, Germany.

Murry, R., and Kothe, E. 2016. Proteomics of inositol phosphate signaling

in the basidiomycete *S. commune. Vereinigung für Allgemeine und Angewandte Mikrobiologie (VAAM) Kongress,* Jena, Germany.

Murry, R., and Kothe, E. 2017. Inositol phosphate signaling in the basidiomycete *Schizophyllum commune*. *13th European Conference on Fungal Genetics,* Paris, France.

# Acknowledgement

I would like to express my gratitude first of all to Prof. Dr. Erika Kothe for the opportunity to work in her working group and laboratory and to accomplish my Ph.D project. I am very grateful for her supervision and support during the Ph.D project. I would never be able to finish without her encouragements and advices that she was constantly giving during the project. I have also to thank her for helpful and fruitful discussions and for her patience and understanding during the time I work for the project. I would also like to thank for many opportunities she gave me such as to do a research stay in London at Prof. Saiardi laboratory, to do an internship at a Biotechnology company Jena Bioscience, and chances to attend several international conferences.

I would like to thank Prof. Axel Brakhage for his second supervision and I would like to thank the ILRS Graduate School for three years of Ph.D project funding. Special thanks go to the ILRS coordinator Dr. Christine Vogler and the former JSMC manager Dr. Carsten Thoms for their help during the recruitment process and everything related to the administration.

I would particularly like to thank Prof. Saiardi for the help and his allowance to do a short-term research stay at his lab at LMCB - MRC Laboratory for Molecular Cell Biology, University College London, UK. Without his guidance and persistent help of writing a manuscript. it would not have been possible to finish. I received generous support from his working group member Cristina Azevedo, Miranda Wilson, Yann Desfougères, Felix and Paloma.

I am thankful for technical support and help of the following people and institutions: Olaf Kniemeyer, Maria Pötsch and Thomas Krüger (Hans Knöll Institute Jena). Petra Mitscherlich (Institue of Microbiology) for the technical assistance in the lab, and Elke for the microscope assistance and small things. I can always ask Elke for anything!

I am particularly thankful for her help regarding administrative issues to our secretary. Christin Reichmann. Special thanks go to Sophia, Julia, Jessi, Olga, Manu, Tosin, Lea, and Nina for the beautiful time and funny moment during hangouts and coffee breaks, but also to the entire Basidiomycete and Microbial communications group for the great working atmosphere. For reading of this thesis and manuscripts I must acknowledge Katrin, Fazil, and Tosin.

For the weekend cool group Sara, Felix, Pol, Lore, Fede, Fernando, Matthias, Beng, and Sindy. I am so happy that you guys exist in this world. Thank you that you are always available to cheer me up. I will never forget stupid but happy moments with you.

At last, I am very grateful for my lovely family, Mama, Papa, Aa, Mas, Kakak, and Ade also the closest friends, who always pray for me, take care of me, have trust in me and motivate me endlessly. I have had the greatest support, motivation, encouragement during ups and downs of this Ph.D project from my beloved Fazil. I can’t imagine doing all of this without you.