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Marc-André Lachance, Editor
University of Western Ontario, London, Ontario, Canada N6A 5B7
<lachance@uwo.ca>

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Editorial

Forthcoming Change in the Editorial Board

Dr. Peter Biely, of the Institute of Chemistry, Slovak Academy of Science, Bratislava, has decided to retire as Associate Editor of the Yeast Newsletter, after 24 years of service. He took over in that role from Dr. Anna Kocková -Kratochvilová in 1991. Peter has assiduously contributed reports on the Annual Meeting of the Czech and Slovak Commission on Yeasts (http://yeastconference.sk/archive), the organization that eventually gave birth to the International Commission on Yeasts, of which the Yeast Newsletter is the official publication. On behalf of our readers, I thank Peter for all he has done over the years and wish him the best in his new endeavours.

MA Lachance, Editor

I Russian Collection of Microorganisms (VKM), Institute for Biochemistry and Physiology of Microorganisms, Pushchino, 142290, Russia - http://www.vkm.ru.
Communicated by WI Golubev <wig@ibpm.pushchino.ru>.

The following paper was recently accepted.

1 Golubev WI 2016 Taxonomic specificity of sensitivity to fungistatic mycocin of *Wickerhamomyces bovis.* Mikrobiologia (Moscow)

Type strain of *Wickerhamomyces bovis* secretes fungistatic mycocin expressed activity within pH range of 3.5 - 6.0. It is most active at pH 5.0 in the presence of 3% NaCl. Yeast species sensitive to this mycocin are grouped in the family Wickerhamomycetaceae and phylogenetically related genera: *Ambrosiozyma, Nakazawaea, Ogataea, Peterozyma*.

The following paper was recently submitted for publication.

2 Golubev W.I. Antifungal activity of methylotrophic yeasts associated with bark beetles. Mykologia i Fitopathologia (Saint-Petersburg).

The strain of *Ogataea pini* was revealed to secrete fungicidal mycocin most active at pH 4.5. It is thermolabile and has small molecular mass. Basidiomycetes are resistant to this mycocin whereas many representatives of the order Saccharomycetales are sensitive to it. The majority of methylotrophic yeasts are resistant or weakly sensitive to this mycocin.

II Biology Department, Brooklyn College, Brooklyn, New York 11210. Communicated by Nasim A. Khan <nasim.khan4@verizon.net>.

Letter to the Editor

The role of mitochondria and IMA genes in α-methylglucoside fermentation in strain 1403-7A of *Saccharomyces cerevisiae*

Strain 1403-7A is constitutive for maltase (E.C. 3.2.1.20) but not for isomaltase (E.C. 3.2.1.10). It is not known if this strain carries any of the IMA genes (IMA1-IMA5) identified by Teste et al. (2010), and by Naumoff and Naumov (2010). This strain ferments maltose and sucrose rapidly but is a slow fermenter of α-methylglucoside. This could be due to slower uptake of alpha-methylglucoside within the cell. We have shown previously the functional mitochondria are required (Khan and Greener 1977) for α-methylglucoside fermentation in strain 1403-7A. When cytoplasmic petites are isolated from this strain after ethidium bromide treatment, they lose the ability to ferment α-methylglucoside completely, but retain the ability to ferment maltose and sucrose, although at a slower rate. Furthermore, when such petites are crossed to a non-fermenting strain, diploids isolated restore the ability to ferment α-methylglucoside as well. These results clearly show α-methylglucoside fermentation in this strain requires the presence of functional mitochondria, and it is an energy requiring processes. Strain 1403-7A is known to carry the MGL3 gene but it is not known if it is allelic to one of the IMA genes.

Teste MA, Francois JM, & Parrou JL 2010 Characterization of a new multigene family encoding isomaltases in the yeast *Saccharomyces cerevisiae*, the IMA family. J Biol Chem 285: 26815-26824.

Naumoff DG & Naumov GI 2010 Discovery of a novel family alpha-glucosidase IMA genes in the yeast *Saccharomyces cerevisiae*, Dokldy Biochemistry and Biophysics, 432(1): 114-116.

Khan NA & Greener A 1977 Effect of the petite mutation on maltose and α-methylglucoside fermentation in *Saccharomyces cerevisiae*. Molec Gen Genet 150:107-108.

III Laboratory of Genetics, Genome Center of Wisconsin, DOE Great Lakes Bioenergy Research Center, Wisconsin Energy Institute, J.F. Crow Institute for the Study of Evolution, University of Wisconsin, Madison, WI 53706, USA. Communicated by Chris Todd Hittinger <cthittinger@wisc.edu>.

Recent publications.

1 Hittinger CT, Rokas A, Bai FY, Boekhout T, Gonçalves P, Jeffries TW, Kominek J, Lachance MA, Libkind D, Rosa CA, Sampaio JP, Kurtzman CP 2015 Genomics and the making of yeast biodiversity. Curr Opin Genet Dev 35:100-109.

Yeasts are unicellular fungi that do not form fruiting bodies. Although the yeast lifestyle has evolved multiple times, most known species belong to the subphylum Saccharomycotina (syn. Hemiascomycota, hereafter yeasts). This diverse group includes the premier eukaryotic model system, *Saccharomyces cerevisiae*; the common human commensal and opportunistic pathogen, *Candida albicans*; and over 1,000 other known species (with more continuing to be discovered). Yeasts are found in every biome and continent and are more genetically diverse than angiosperms or chordates. Ease of culture, simple life cycles, and small genomes (~10-20 Mbp) have made yeasts exceptional models for molecular genetics, biotechnology, and evolutionary genomics. Here we discuss recent developments in understanding the genomic underpinnings of the making of yeast biodiversity, comparing and contrasting natural and human-associated evolutionary processes. Only a tiny fraction of yeast biodiversity and metabolic capabilities has been tapped by industry and science. Expanding the taxonomic breadth of deep genomic investigations will further illuminate how genome function evolves to encode their diverse metabolisms and ecologies.

2 Alexander WG, Peris D, Pfannenstiel BT, Opulente DA, Kuang M, Hittinger CT 2016 Efficient engineering of marker-free synthetic allotetraploids of *Saccharomyces*. Fungal Genet Biol 89:10-17.

Saccharomyces interspecies hybrids are critical biocatalysts in the fermented beverage industry, including in the production of lager beers, Belgian ales, ciders, and cold-fermented wines. Current methods for making synthetic interspecies hybrids are cumbersome and/or require genome modifications. We have developed a simple, robust, and efficient method for generating allotetraploid strains of prototrophic *Saccharomyces* without sporulation or nuclear genome manipulation. *S. cerevisiae x S. eubayanus*, *S. cerevisiae x S. kudriavzevii*, and *S. cerevisiae x S. uvarum* designer hybrid strains were created as synthetic lager, Belgian, and cider strains, respectively. The ploidy and hybrid nature of the strains were confirmed using flow cytometry and PCR-RFLP analysis, respectively. This method provides an efficient means for producing novel synthetic hybrids for beverage and biofuel production, as well as for constructing tetraploids to be used for basic research in evolutionary genetics and genome stability.

3 Leducq JB, Nielly-Thibault L, Charron G, Eberlein C, Verta JP, Samani P, Sylvester K, Hittinger CT, Bell G, Landry CR 2016 Speciation driven by hybridization and chromosomal plasticity in a wild yeast. Nat Microbiol 1:15003.

Hybridization is recognized as a powerful mechanism of speciation and a driving force in generating biodiversity. However, only few multicellular species, limited to a handful of plants and animals, have been shown to fulfill all the criteria of homoploid hybrid speciation. This lack of evidence could lead to the interpretation that speciation by hybridization has a limited role in eukaryotes, particularly in single-celled organisms. Laboratory experiments have revealed that fungi such as budding yeasts can rapidly develop reproductive isolation and novel phenotypes through hybridization, showing that in principle homoploid speciation could occur in nature. Here, we report a case of homoploid hybrid speciation in natural populations of the budding yeast *Saccharomyces paradoxus* inhabiting the North American forests. We show that the rapid evolution of chromosome architecture and an ecological context that led to secondary contact between nascent species drove the formation of an incipient hybrid species with a potentially unique ecological niche.

4 Alexander WG, Wisecaver JH, Rokas A, Hittinger CT 2016 Horizontally acquired genes in early-diverging pathogenic fungi enable the use of host nucleosides and nucleotides. Proc Natl Acad Sci USA 113:4116-21.

Horizontal gene transfer (HGT) among bacteria, archaea, and viruses is widespread, but the extent of transfers from these lineages into eukaryotic organisms is contentious. Here we systematically identify hundreds of genes that were likely acquired horizontally from a variety of sources by the early-diverging fungal phyla Microsporidia and Cryptomycota. Interestingly, the Microsporidia have acquired via HGT several genes involved in nucleic acid synthesis and salvage, such as those encoding thymidine kinase (*TK*), cytidylate kinase, and purine nucleotide phosphorylase. We show that these HGT-derived nucleic acid synthesis genes tend to function at the interface between the metabolic networks of the host and pathogen. Thus, these genes likely play vital roles in diversifying the useable nucleic acid components available to the intracellular parasite, often through the direct capture of resources from the host. Using an in vivo viability assay, we also demonstrate that one of these genes, *TK*, encodes an enzyme that is capable of activating known prodrugs to their active form, which suggests a possible treatment route for microsporidiosis. We further argue that interfacial genes with well-understood activities, especially those horizontally transferred from bacteria or viruses, could provide medical treatments for microsporidian infections.

5 Wisecaver JH, Alexander WG, King SB, Hittinger CT, Rokas A 2016 Dynamic evolution of nitric oxide detoxifying flavohemoglobins, a family of single-protein metabolic modules in bacteria and eukaryotes. Mol Biol Evol (in press) - doi: 10.1093/molbev/msw073

Due to their functional independence, proteins that comprise standalone metabolic units, which we name single-protein metabolic modules, may be particularly prone to gene duplication (GD) and horizontal gene transfer (HGT). Flavohemoglobins (flavoHbs) are prime examples of single-protein metabolic modules, detoxifying nitric oxide (NO), a ubiquitous toxin whose antimicrobial properties many life forms exploit, to nitrate, a common source of nitrogen for organisms. FlavoHbs appear widespread in bacteria and have been identified in a handful of microbial eukaryotes, but how the distribution of this ecologically and biomedically important protein family evolved remains unknown. Reconstruction of the evolutionary history of 3,318 flavoHb protein sequences covering the family's known diversity showed evidence of recurrent HGT at multiple evolutionary scales including intra-bacterial HGT, as well as HGT from bacteria to eukaryotes. One of the most striking examples of HGT is the acquisition of a flavoHb by the dandruff- and eczema-causing fungus *Malassezia* from *Corynebacterium* Actinobacteria, a transfer that growth experiments show is capable of mediating NO resistance in fungi. Other flavoHbs arose via GD; for example, many filamentous fungi possess two flavoHbs that are differentially targeted to the cytosol and mitochondria, likely conferring protection against external and internal sources of NO, respectively. Because single-protein metabolic modules such as flavoHb function independently, readily undergo GD and HGT, and are frequently involved in organismal defense and competition, we suggest that they represent "plug-and-play" proteins for ecological arms races.

6 McIlwain SJ, Peris D, Sardi M, Moskvin OV, Zhan F, Myers K, Riley NM, Buzzell A, Parreiras L, Ong IM, Landick R, Coon JJ, Gasch AP, Sato TK, Hittinger CT 2016 Genome sequence and analysis of a stress-tolerant, wild-derived strain of *Saccharomyces cerevisiae* used in biofuels research. G3 (Bethesda) (in press) - doi: 10.1534/g3.116.029389

The genome sequences of more than 100 strains of the yeast *Saccharomyces cerevisiae* have been published. Unfortunately, most of these genome assemblies contain dozens to hundreds of gaps at repetitive sequences, including transposable elements, tRNAs, and subtelomeric regions, which is where novel genes generally reside. Relatively few strains have been chosen for genome sequencing based on their biofuel production potential, leaving an additional knowledge gap. Here we describe the nearly complete genome sequence of GLBRCY22-3 (Y22-3), a strain of *S. cerevisiae* derived from the stress-tolerant wild strain NRRL YB-210 and subsequently engineered for xylose metabolism. After

benchmarking several genome assembly approaches, we developed a pipeline to integrate Pacific Biosciences (PacBio) and Illumina sequencing data and achieved one of the highest quality genome assemblies for any *S. cerevisiae* strain. Specifically, the contig N50 is 693 kbp, and the sequences of most chromosomes, the mitochondrial genome, and 2-micron plasmid are complete. Our annotation predicts 92 genes that are not present in the reference genome of the laboratory strain S288c, over 70% of which were expressed. We predicted functions for 43 of these genes, 28 of which were previously uncharacterized and unnamed. Remarkably, many of these genes are predicted to be involved in stress tolerance and carbon metabolism and are shared with a Brazilian bioethanol production strain, even though the strains differ dramatically at most genetic loci. The Y22-3 genome sequence provides an exceptionally high-quality resource for basic and applied research in bioenergy and genetics.

7 Lopes MR, Morais CG, Kominek J, Cadete RM, Soares MA, Uetanabaro APT, Fonseca C, Lachance MA, Hittinger CT, Rosa CA 2016 Genomic analysis and D-xylose fermentation of three novel *Spathaspora* species: *Spathaspora girioi* sp. nov., *Spathaspora hagerdaliae* f. a., sp. nov., and *Spathaspora gorwiae* f. a., sp. nov. FEMS Yeast Res (in press) - doi: 10.1093/femsyr/fow044

Three novel D-xylose-fermenting yeast species of *Spathaspora* clade were recovered from rotting wood in regions of the Atlantic Rainforest ecosystem in Brazil. Differentiation of new species was based on analyses of the gene encoding the D1/D2 sequences of large subunit of rRNA and on 642 conserved, single-copy, orthologous genes from genome sequence assemblies from the newly described species and 15 closely-related Debaryomycetaceae / Metschnikowiaceae species. *Spathaspora girioi* sp. nov. produced unconjugated asci with a single elongated ascospore with curved ends; ascospore formation was not observed for the other two species. The three novel species ferment D-xylose with different efficiencies. *Spathaspora hagerdaliae* sp. nov. and *Spathaspora girioi* sp. nov. showed xylose reductase (XR) activity strictly dependent on NADPH, whereas *Spathaspora gorwiae* sp. nov. had XR activity that used both NADH and NADPH as co-factors. The genes that encode enzymes involved in D-xylose metabolism (xylose reductase, xylitol dehydrogenase, and xylulokinase) were also identified for these novel species. The type strains are *Spathaspora girioi* sp. nov. UFMG-CM-Y302^T (= CBS 13476), *Spathaspora hagerdaliae* f.a., sp. nov. UFMG-CM-Y303^T (= CBS 13475), and *Spathaspora gorwiae* f.a., sp. nov. UFMG-CM-Y312^T (= CBS 13472).

IV VTT Technical Research Centre of Finland, P.O. Box 1000, FI-02044 VTT, Finland. Communicated by Brian Gibson <brian.gibson@vtt.fi>.

Recent publications.

1 Rantasalo A, Czeizler E, Virtanen R, Rousu J, Lähdesmäki H, Penttilä M, Jäntti J, Mojzita D 2016 Synthetic transcription amplifier system for orthogonal control of gene expression in *Saccharomyces cerevisiae*. PLoS One. 11(2):e0148320 - doi: 10.1371/journal.pone.0148320

This work describes the development and characterization of a modular synthetic expression system that provides a broad range of adjustable and predictable expression levels in *S. cerevisiae*. The system works as a fixed-gain transcription amplifier, where the input signal is transferred via a synthetic transcription factor (sTF) onto a synthetic promoter, containing a defined core promoter, generating a transcription output signal. The system activation is based on the bacterial LexA-DNA-binding domain, a set of modified, modular LexA-binding sites and a selection of transcription activation domains. We show both experimentally and computationally that the tuning of the system is achieved through the selection of three separate modules, each of which enables an adjustable output signal: 1) the transcription-activation domain of the sTF, 2) the binding-site modules in the output promoter, and 3) the core promoter modules which define the transcription initiation site in the output promoter. The system has a novel bidirectional architecture that enables generation of compact, yet versatile expression modules for multiple genes with highly diversified expression levels ranging from negligible to very strong using one synthetic transcription factor. In contrast to most existing modular gene expression regulation systems, the

present system is independent from externally added compounds. Furthermore, the established system was minimally affected by the several tested growth conditions. These features suggest that it can be highly useful in large scale biotechnology applications.

2 Weber-Boyvat M, Chernov KG, Aro N, Wohlfahrt G, Olkkonen VM, Jäntti J 2016 The Sec1/Munc18 protein groove plays a conserved role in interaction with Sec9p/SNAP-25. Traffic. 2016 Feb;17(2):131-53. doi: 10.1111/tra.12349.

The Sec1/Munc18 (SM) proteins constitute a conserved family with essential functions in SNARE-mediated membrane fusion. Recently, a new protein–protein interaction site in Sec1p, designated the groove, was proposed. Here, we show that a *sec1* groove mutant yeast strain, *sec1(w24)*, displays temperature-sensitive growth and secretion defects. The yeast Sec1p and mammalian Munc18-1 grooves were shown to play an important role in the interaction with the SNAREs Sec9p and SNAP-25b, respectively. Incubation of SNAP-25b with the Munc18-1 groove mutant resulted in a lag in the kinetics of SNARE complex assembly *in vitro* when compared with wild-type Munc18-1. The SNARE regulator *SRO7* was identified as a multicopy suppressor of *sec1(w24*) groove mutant and an intact Sec1p groove was required for the plasma membrane targeting of Sro7p–SNARE complexes. Simultaneous inactivation of Sec1p groove and *SRO7* resulted in reduced levels of exocytic SNARE complexes. Our results identify the groove as a conserved interaction surface in SM proteins. The results indicate that this structural element is important for interactions with Sec9p/SNAP-25 and participates, in concert with Sro7p, in the initial steps of SNARE complex assembly.

3 Weber-Boyvat M, Li S, Skarp KP, Olkkonen VM, Yan D, Jäntti J 2015 Bimolecular fluorescence complementation (BiFC) technique in yeast *Saccharomyces cerevisiae* and mammalian cells. Methods Mol Biol 1270:277-88 - doi: 10.1007/978-1-4939-2309-0_20.

Visualization of protein–protein interactions in vivo offers a powerful tool to resolve spatial and temporal aspects of cellular functions. The bimolecular fluorescence complementation (BiFC) makes use of nonfluorescent fragments of green fluorescent protein or its variants that are added as “tags” to target proteins under study. Only upon target protein interaction is a fluorescent protein complex assembled, and the site of interaction can be monitored by microscopy. In this chapter, we describe the method and tools for the use of BiFC in the yeast *Saccharomyces cerevisiae* and in mammalian cells.

4 Krogerus K, Arvas M, De Chiara M, Mattinen L, Magalhães F, Oja M, Yue JX, Liti G. & Gibson B 2016 Ploidy influences the functional attributes of *de novo* lager yeast hybrids. Appl Microbiol Biotechnol - DOI: 10.1007/s00253-016-7588-3

The genomes of hybrid organisms, such as lager yeast (Saccharomyces cerevisiae × *Saccharomyces eubayanus*), contain orthologous genes, the functionality and effect of which may differ depending on their origin and copy number. How the parental subgenomes in lager yeast contribute to important phenotypic traits such as fermentation performance, aroma production, and stress tolerance remains poorly understood. Here, three de novo lager yeast hybrids with different ploidy levels (allodiploid, allotriploid, and allotetraploid) were generated through hybridization techniques without genetic modification. The hybrids were characterized in fermentations of both high gravity wort (15 °P) and very high gravity wort (25 °P), which were monitored for aroma compound and sugar concentrations. The hybrid strains with higher DNA content performed better during fermentation and produced higher concentrations of flavor-active esters in both worts. The hybrid strains also outperformed both the parent strains. Genome sequencing revealed that several genes related to the formation of flavor-active esters (ATF1, ATF2, EHT1, EEB1, and BAT1) were present in higher copy numbers in the higher ploidy hybrid strains. A direct relationship between gene copy number and transcript level was also observed. The measured ester concentrations and transcript levels also suggest that the functionality of the S. *cerevisiae*- and S. *eubayanus*-derived gene products differs. The results contribute to our understanding of the complex molecular mechanisms that determine phenotypes in lager yeast hybrids and are expected to facilitate targeted strain development through interspecific hybridization.

5 Nathanail AV, Gibson B, Han L, Peltonen K, Ollilainen V, Jestoi M & Laitila A 2016 The lager yeast *Saccharomyces pastorianus* removes and transforms *Fusarium trichothecene* mycotoxins during fermentation of brewer's wort. Food Chem 203:448-455.

An investigation was conducted to determine the fate of deoxynivalenol, deoxynivalenol-3-glucoside, HT-2 toxin and T-2 toxin, during a four-day fermentation with the lager yeast *Saccharomyces pastorianus*. The influence of excessive mycotoxin concentrations on yeast growth, productivity and viability were also assessed. Mycotoxins were dosed at varying concentrations to 11.5° Plato wort. Analysis of yeast revealed that presence of the toxins even at concentrations up to 10,000 µg/L had little or no effect on sugar utilisation, alcohol production, pH, yeast growth or cell viability. Of the dosed toxin amounts 9–34% were removed by the end of fermentation, due to physical binding and/or biotransformation by yeast. Deoxynivalenol-3-glucoside was not reverted to its toxic precursor during fermentation. Processing of full-scan liquid chromatography-quadrupole time-of-flight-mass spectrometry (LC–QTOF–MS) data with MetaboLynx™ and subsequent LC–QTOF–MS/MS measurements resulted in annotation of several putative metabolites. De(acetylation), glucosylation and sulfonation were the main metabolic pathways activated.

V Yeast Molecular Genetics Laboratory, Institute of Molecular Biology "Acad. Rumen Tsanev", Bulgarian Academy of Sciences, Acad. G. Bonchev str., 1113 Sofia, Bulgaria. Communicated by George Miloshev <miloshev@bio21.bas.bg> - http://www.chromatinepigenetics.com

The following is the abstract of a recently published paper of the group.

1 Staneva D, Georgieva M, Miloshev G 2016 *Kluyveromyces lactis* genome harbours a functional linker histone encoding gene. FEMS Yeast Res 16(4) - doi: 10.1093/femsyr/fow034.

Linker histones are essential components of chromatin in eukaryotes. Through interactions with linker DNA and nucleosomes they facilitate folding and maintenance of higher-order chromatin structures and thus delicately modulate gene activity. The necessity of linker histones in lower eukaryotes appears controversial and dubious. Genomic data have shown that *Schizosaccharomyces pombe* does not possess genes encoding linker histones while *Kluyveromyces lactis* has been reported to have a pseudogene. Regarding this controversy, we have provided the first direct experimental evidence for the existence of a functional linker histone gene, *KlLH1*, in *K. lactis* genome. Sequencing of *KlLH1* from both genomic DNA and copy DNA confirmed the presence of an intact open reading frame. Transcription and splicing of the *KlLH1* sequence as well as translation of its mRNA have been studied. *In silico* analysis revealed homology of KlLH1p to the histone H1/H5 protein family with predicted three domain structure characteristic for the linker histones of higher eukaryotes. This strongly proves that the yeast *K. lactis* does indeed possess a functional linker histone gene thus entailing the evolutionary preservation and significance of linker histones. The nucleotide sequences of *KlLH1* are deposited in the GenBank under accession numbers KT826576, KT826577 and KT826578.

VI Department of AGRARIA, "*Mediterranea*" University of Reggio Calabria, Via Feo di Vito, I-89122 Reggio Calabria, Italy. Communicated by Andrea Caridi <acaridi@unirc.it>.

Recent publication.

1 Sidari R, Caridi A 2016 Nutrient depletion modifies cell wall adsorption activity of wine yeast. World J Microbiol Biotechnol - DOI 10.1007/s11274-016-2047-y

Yeast cell wall is a structure that helps yeasts to manage and respond to many environmental stresses. The mannosyl-phosphorylation is a modification in response to stress that provides the cell wall with negative charges able to bind compounds present in the environment. Phenotypes related to the cell wall modification such as the filamentous growth in *Saccharomyces cerevisiae* are affected by nutrient depletion. The present work aimed at describing the effect of carbon and/or nitrogen limitation on the aptitude of *S. cerevisiae* strains to bind coloured polyphenols. Carbon- and nitrogen-rich or deficient

media supplemented with grape polyphenols were used to simulate different grape juice conditions - early, mid, 'adjusted' for nitrogen, and late fermentations. In early fermentation condition, the R+G+B values range from 106 (high adsorption, strain Sc1128) to 192 (low adsorption, strain R1278b), in mid-fermentation the values range from 111 (high adsorption, strain Sc1321) to 258 (low adsorption, strain Sc2306), in 'adjusted' for nitrogen conditions the values range from 105 (high adsorption, strain Sc1321) to 194 (low adsorption, strain Sc2306) while in late fermentation conditions the values range from 101 (high adsorption, strain Sc384) to 293 (low adsorption, strain Sc2306). The effect of nutrient availability is not univocal for all the strains and the different media tested modified the strains behaviour. In all the media the strains show significant differences. Results demonstrate that wine yeasts decrease/increase their parietal adsorption activity according to the nutrient availability. The wide range of strain variability observed could be useful in selecting wine starters.

VII Laboratory of Genetics, Genome Center of Wisconsin, DOE Great Lakes Bioenergy Research Center, Wisconsin Energy Institute, University of Wisconsin, Madison, WI 53706, USA. Communicated by David Peris <david.perisnavarro@wisc.edu>.

Recent publication.

1 Peris D, Arias A, Orlić S, Belloch C, Pérez-Través L, Querol A, Barrio E Mitochondrial introgression suggests extensive ancestral hybridization events among *Saccharomyces* species - BioRχiv - doi: http://dx.doi.org/10.1101/028324.

Horizontal Gene Transfer (HGT) in eukaryotic plastids and mitochondrial genomes is frequently observed, and plays an important role in organism evolution. In yeasts, recent mitochondrial HGT has been suggested between *S. cerevisiae* and *S. paradoxus*. However, few strains have been explor-ed due to the lack of accurate mitochondrial genome annotations. Mitochondrial genome sequences are important to understand how frequent these introgressions occur and their role in cytonuclear incompatibilities. In fact, most of the Bateson-Dobzhansky-Muller genetic incompatibilities described in yeasts are driven by these cytonuclear incompatibilities. In this study, we have explored the mitochondrial inheritance of several worldwide distributed *Saccharomyces* species isolated from different sources and geographic origins. We demonstrated the existence of recombination hotspots in the mitochondrial region *COX2-ORF1*, likely mediated by the transfer of two different types of *ORF1*, encoding a free-standing homing endonuclease, or facilitated by AT tandem repeats and GC clusters. These introgressions were shown to occur both at intra- and interspecific levels. Based on our results we proposed a model which involve several ancestral hybridization events among *Saccharomyces* strains in wild environments.

VIII Bioprocess & Metabolic Engineering Lab (LEMeB), University of Campinas, Campinas SP, Brazil. Commuicated by Andreas K. Gombert <gombert@unicamp.br>.

We recently published a mini-review on the metabolism of sucrose in *Saccharomyces cerevisiae*:

1 Marques WL, Raghavendran V, Stambuk BU, Gombert AK 2016 Sucrose and *Saccharomyces cerevisiae*: a relationship most sweet. FEMS Yeast Res 16 - doi: 10.1093/femsyr/fov107

Sucrose is an abundant, readily available and inexpensive substrate for industrial biotechnology processes and its use is demonstrated with much success in the production of fuel ethanol in Brazil. *Saccharomyces cerevisiae*, which naturally evolved to efficiently consume sugars such as sucrose, is one of the most important cell factories due to its robustness, stress tolerance, genetic accessibility, simple nutrient requirements and long history as an industrial workhorse. This minireview is focused on sucrose metabolism in *S. cerevisiae*, a rather unexplored subject in the scientific literature. An analysis of sucrose availability in nature and yeast sugar metabolism was performed, in order to understand the molecular background that makes *S. cerevisiae* consume this sugar efficiently. A historical overview on the use of sucrose and *S. cerevisiae* by humans is also presented considering sugarcane and sugarbeet as the main sources of this carbohydrate. Physiological aspects of sucrose consumption are compared with

those concerning other economically relevant sugars. Also, metabolic engineering efforts to alter sucrose catabolism are presented in a chronological manner. In spite of its extensive use in yeast-based industries, a lot of basic and applied research on sucrose metabolism is imperative, mainly in fields such as genetics, physiology and metabolic engineering.

IX CBS-KNAW Fungal Biodiversity Centre, Uppsalalaan 8, 3584 CT, Utrecht, The Netherlands. Communicated by Marizeth Groenewald <m.groenewald@cbs.knaw.nl>.

Recent publications.

1 Gouliamova DE, Dimitrov RA, Smith MTh, Groenewald M, Stoilova-Disheva MM, Gueorguiev BV, Boekhout T 2016 DNA barcoding revealed *Nematodospora valgi* gen. nov., sp. nov. and *Candida cetoniae* sp. nov. in the Lodderomyces clade. Fungal Biology 120: 179–190.

2 Gouliamova, DE, Dimitrov RA, Smith MTh, Groenewald M, Stoilova-Disheva MM., Boekhout T 2016 Fungal Planet description sheets: 320–370: *Metschnikowia colchici* DE Gouliamova, RA Dimitrov, MT Sm., M Groenew., MM Stoilova-Disheva & Boekhout, *sp. nov.* Persoonia 34:167–266.

3 Liu X, Wang QM, Theelen B, Groenewald M, Bai FY, Boekhout T 2016 Phylogeny of tremellomycetous yeasts reconstructed from multiple gene sequence analyses. Studies in Mycology 81: 1-26.

4 Wang QM, Groenewald M, Takashima M, Theelen B, Han PJ, Liu XZ, Boekhout T, Bai, F.-Y. 2016. Phylogeny of yeasts and related taxa within Pucciniomycotina determined from multigene gene sequence analyses. Studies in Mycology 81:27–53.

5 Wang QM, Begerow, D., Groenewald M, Liu X-Z., Theelen B, Bai FY, Boekhout T 2016 Multigene phylogeny and taxonomic revision of yeasts and related fungi in the Ustilaginomycotina Studies in Mycology 81: 55–83.

6 Liu XZ, Wang QM, Göker M, Groenewald M, Kachalkin AV, Lumbsch HT, Millanes, Wedin AMM, Yurkov AM, Boekhout T, Bai FY 2016. Towards an integrated phylogenetic classification of the Tremellomycetes. Studies in Mycology 81: 85–147.

7 Wang QM, Yurkov, A. Göker M M, Lumbsch, H. T., Leavitt S D., Groenewald M, Theelen B, Liu X-Z., Boekhout T, Bai FY 2016. Phylogenetic classification of yeasts and related taxa within Pucciniomycotina. Studies in Mycology 81:149–189.

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XI Department of Medical Microbiology and Infectious Diseases, Canisius-Wilhelmina Hospital, Nijmegen, The Netherlands. Communicated by Ferry Hagen <F.Hagen@cwz.nl>.

Recent publications.

1 Hagen F, Hare Jensen R, Meis JF, Arendrup MC 2016 Molecular epidemiology and *in vitro* antifungal susceptibility testing of 108 clinical *Cryptococcus neoformans sensu lato* and *Cryptococcus gattii sensu lato* isolates from Denmark. Mycoses in press - doi: 10.1111/myc.12507.

Cryptococcosis is mainly caused by members of the *Cryptococcus gattii*/*Cryptococcus neoformans* species complexes. Here, we report the molecular characterisation and in vitro antifungal susceptibility of Danish clinical cryptococcal isolates. Species, genotype, serotype and mating type were determined by amplified fragment length polymorphism (AFLP) fingerprinting and qPCR. EUCAST E.Def 7.2 MICs were determined for amphotericin B, flucytosine, fluconazole, voriconazole and isavuconazole. Most isolates were *C. neoformans* (serotype A; *n* = 66) and belonged to genotype AFLP1/VNI (*n* = 61) or AFLP1B/VNII (*n* = 5) followed by *Cryptococcus deneoformans* (serotype D; genotype AFLP2, *n* = 20), *C. neoformans* × *C. deneoformans* hybrids (serotype AD; genotype AFLP3, *n* = 13) and *Cryptococcus curvatus* (*n* = 2). Six isolates were *C. gattii sensu lato*, and one isolate was a *C. deneoformans* × *C. gattii* hybrid (genotype AFLP8). All isolates were amphotericin B susceptible. Flucytosine susceptibility was uniform MIC_{50} of 4-8 mg l^{-1} except for *C. curvatus* (MICs >32 mg l^{-1}). *Cryptococcus gattii sensu lato* isolates were somewhat less susceptible to the azoles. MICs of fluconazole (>32 mg l^{-1}), voriconazole (≥0.5 mg l^{-1}) and isavuconazole (0.06 and 0.25 mg l^{-1} respectively) were elevated compared to the wild-type population for 1/19 *C. deneoformans* and 1/2 *C. curvatus* isolates. Flucytosine MIC was elevated for 1/61 *C. neoformans* (>32 mg l^{-1}). Antifungal susceptibility revealed species-specific differential susceptibility, but suggested acquired resistance was an infrequent phenomenon.

2 Nyazika TK, Hagen F, Meis JF, Robertson VJ 2016 *Cryptococcus tetragattii* as a major cause of cryptococcal meningitis among HIV-infected individuals in Harare, Zimbabwe. J Infect (in press) - doi: 10.1016/j.jinf.2016.02.018.

HIV-associated cryptococcal meningitis is commonly caused by *Cryptococcus neoformans*, whilst infections with *Cryptococcus gattii sensu lato* are historically rare. Despite available studies, little is known about the occurrence of *C. gattii sensu lato* infections among HIV-infected individuals in Zimbabwe. In a prospective cohort, we investigated the prevalence of *C. gattii sensu lato* meningitis among HIV-infected patients (*n* = 74) in Harare, Zimbabwe. Of the 66/74 isolates confirmed by molecular characterization, 16.7% (11/66) were found to be *C. gattii sensu lato* and 83.3% (55/66) *C. neoformans sensu stricto*. From one patient two phenotypically different *C. gattii sensu lato* colonies were cultured. The majority (*n* = 9/12; 75%) of the *C. gattii sensu lato* isolates were *Cryptococcus tetragattii* (AFLP7/VGIV), which has been an infrequently reported pathogen. In-hospital mortality associated with *C. gattii sensu lato* was 36.4%. Our data suggests that *C. tetragattii* (AFLP7/VGIV) is a more common cause of disease than *C. gattii sensu stricto* (genotype AFLP4/VGI) among patients with HIV-associated cryptococcal meningitis in Harare, Zimbabwe and possibly underreported in sub-Saharan Africa.

3 Nyazika TK, Robertson VJ, Nherera B, Mapondera PT, Meis JF, Hagen F 2016 Comparison of biotyping methods as alternative identification tools to molecular typing of pathogenic *Cryptococcus* species in sub-Saharan Africa. Mycoses. 59(3):151-156 - doi: 10.1111/myc.12444.

Cryptococcal meningitis is the leading fungal infection and AIDS defining opportunistic illness in patients with late stage HIV infection, particularly in South-East Asia and sub-Saharan Africa. Given the high mortality, clinical differences and the extensive ecological niche of *Cryptococcus neoformans* and *Cryptococcus gattii* species complexes, there is need for laboratories in sub-Sahara African countries to adopt new and alternative reliable diagnostic algorithms that rapidly identify and distinguish these species. We biotyped 74 and then amplified fragment length polymorphism (AFLP) genotyped 66 *Cryptococcus* isolates from a cohort of patients with HIV-associated cryptococcal meningitis. *C. gattii sensu lato* was isolated at a prevalence of 16.7% (n = 11/66) and *C. neoformans sensu stricto* was responsible for 83.3% (n = 55/66) of the infections. L-Canavanine glycine bromothymol blue, yeast-carbon-base-D-proline-D-tryptophan and creatinine dextrose bromothymol blue thymine were able to distinguish pathogenic *C. gattii sensu lato* from *C. neoformans sensu stricto* species when compared with AFLP genotyping. This study demonstrates high *C. gattii sensu lato* prevalence in Zimbabwe. In addition, biotyping methods can be used as alternative diagnostic tools to molecular typing in resource-limited areas for differentiating pathogenic *Cryptococcus* species.

XII Department of Bacteriology, University of Wisconsin-Madison, Madison, WI 53706, USA. Communicated by Christopher Calvey <calvey@wisc.edu>.

Recent publication.

1 Calvey CH, Su YK, Willis LB, McGee M, Jeffries TW 2016 Nitrogen limitation, oxygen limitation, and lipid accumulation in *Lipomyces starkeyi*. Bioresour Technol 200:780-788 - doi: 10.1016 /j.biortech.2015.10.104.

Lipid production by oleaginous yeasts is optimal at high carbon-to-nitrogen ratios. In the current study, nitrogen and carbon consumption by *Lipomyces starkeyi* were directly measured in defined minimal media with nitrogen content and agitation rates as variables. Shake flask cultures with an initial C:N ratio of 72:1 cultivated at 200rpm resulted in a lipid output of 10g/L, content of 55%, yield of 0.170g/g, and productivity of 0.06g/L/h. All of these values decreased by ≈50-60% when the agitation rate was raised to 300rpm or when the C:N ratio was lowered to 24:1, demonstrating the importance of these parameters. Under all conditions, *L. starkeyi* cultures tolerated acidified media (pH≈2.6) without difficulty, and produced considerable amounts of alcohols; including ethanol, mannitol, arabitol, and 2,3-butanediol. *L. starkeyi* also produced lipids from a corn stover hydrolysate, showing its potential to produce biofuels from renewable agricultural feedstocks.

XIII Department of Soil Biology, Faculty of Soil Science, Lomonosov Moscow State University, 119991, Leninskie gory, 1/12, Moscow, Russia. Communicated by Aleksey Kachalkin <kachalkin_a@mail.ru>.

Recent publications.

1 Kachalkin AV, Abdullabekova DA, Magomedova ES, Magomedov GG, Chernov IYu 2015 Yeasts of the vineyards in Dagestan and other regions. Microbiology 84(3):425-432.

Long-term studies of yeast species diversity in the vineyards of the Republic of Dagestan using various isolation techniques and various substrates in the vertical tier dynamics revealed 38 species. The most diverse species complex including ~80% of the isolated species was formed on the berries. A list of 160 yeast species isolated from grapes, spontaneously fermented fresh juice, and other vineyard substrates was compiled using the results of the present work and the literature data on yeast occurrence. Analysis of generalized data revealed considerable similarity in the taxonomic composition of yeasts from different countries and continents and made it possible to shift from the genus to the species characterization of the

grape-associated yeast community.

2 Glushakova AM, Kachalkin AV, Chernov Iyu 2015 Soil yeast communities under the aggressive invasion of Sosnowsky's hogweed (*Heracleum sosnowskyi*). Eurasian Soil Sci 48(2):201-207.

The year-round dynamics of the number and taxonomic composition of yeast communities in the soddy-podzolic soils under invasive thickets of Heracleum sosnowskyi were investigated. The yeast groups that are formed in the soil under the continuous Sosnowsky's hogweed thickets significantly differ from the indigenous yeast communities under the adjacent meadows. In the soils of both biotopes, typical eurybiotic yeast species predominate. In the soil under Heracleum sosnowskyi, the share of the ascomycetes *Candida vartiovaarae* and *Wickerhamomyces anomalus* is much lower, and the portion of yeast-like fungi with high hydrolytic activity such as *Trichosporon moniliforme* and *Trichosporon porosum* is greater. A possible explanation for this phenomenon is that Sosnowsky's hogweed, unlike most aboriginal meadow grasses, does not hibernate with green leaves that do not gradually die out with the formation of semidecomposed plant residues—the main source of nutrients for the soil-litter microbial complex. In addition, grasses of the lower layer do not develop under Sosnowsky's hogweed due to the strong shading and allelopathic impact preventing the development of typical epiphytic copiotrophic species of yeasts.

3 Glushakova AM, Kachalkin AV, Chernov Iyu 2015 Effect of invasive herb species on the structure of soil yeast complexes in mixed forests exemplified by *Impatiens parviflora* DC. Microbiology 84(5):717-721.

Yeast abundance and diversity in a mixed forest sod–podzol soil under *Impatiens parviflora* DC plants was studied in comparison with unimpaired aboriginal herbaceous plants typical of the Central Russian secondary, after-forest meadow. The study was carried out throughout the vegetation period. Standard microbiological plating techniques revealed 36 yeast species. Typical pedobiotic (*Cryptococcus podzolicus, Wickerhamomyces anomalus*) and eurybiotic yeast species (*Rhodotorula mucilaginosa*) predominated in both biotopes. The relative abundance of the autochthonous soil yeast species *Cryptococcus podzolicus* was higher in the soil under aboriginal herbs than under *Impatiens parviflora*. Sites with aboriginal vegetation were also characterized by high abundance of the pedogamous species *Schwanniomyces castelli* and *Torulaspora delbrueckii*. The share of yeastlike Trichosporon fungi with high hydrolytic activity was considerably higher under adventitious plants *Impatiens parviflora*, as well as in the previously studied soil under *Heracleum sosnowskyi*.

4 Glushakova AM, Kachalkin AV, Zheltikova TM, Chernov Iyu 2015 Resistance of various yeast ecological groups to prolonged storage in dry state. Microbiology 84(3):442-448.

Resistance of 14 yeast species belonging to different ecological groups to extensive storage in a dried state was investigated. Pedobiotic yeasts isolated mainly from the soils of humid areas (*Cryptococcus podzolicus, Cr. terricola,* and *Lipomyces starkeyi*) were the least resistant. The yeasts associated with the nectar of entomophilous plants (*Metschnikowia reukaufii* and *Candida bombi*) also exhibited low resistance to drying. Complete death of these species occurred during the first month of storage. Eurybiotic species from various environments (*Cryptococcus magnus, Cryptococcus victoriae, Debaryomyces hansenii,* and *Cryptococcus wieringae*) were somewhat more resistant. Pigmented plant-associated yeasts (*Rhodotorula mucilaginosa* and *Sporobolomyces roseus*), as well as the pathogenic or opportunistic *Candida* strains (*C. albicans* and *C. parapsilosis*), were the most resistant to drying. Thus, occurrence of yeasts in natural habitats is closely associated with their ability to survive prolonged drying.

5 Maksimova IA, Glushakova AM, Kachalkin AV, Chernov IYu, Panteleeva SN, Reznikova ZhI 2016 Yeast communities of *Formica aquilonia* colonies. Microbiology 85(1):124-129.

Yeast abundance and species diversity in the colonies of *Formica aquilonia* ants in birch–pine grass forest near Novosibirsk, Russia, were studied. The average yeast number in the anthill material was

10^3–10^4 CFU/g, reaching 10^5 CFU/g in the hatching chambers. Typical litter species (*Trichosporon moniliiforme* and *Cystofilobasidium capitatum*) were predominant in soil and litter around the anthills. Apart from these species, ascomycete species of the family *Debaryomycetaceae*, *Debaryomyces hansenii,* and *Schwanniomyces vanrijiae* were predominant in the anthill material. Yeast population of the ant' bodies consisted exclusively of the members of the last two species. Thus, highly specific yeast communities formed in the colonies of *Formica aquilonia* ants differ from the communities of surrounding soil. These differences are caused by environment-forming activity of the ants.

XIV Department of Microbial, Biochemical & Food Biotechnology, University of the Free State, P.O. Box 339, Bloemfontein 9300, South Africa - http://www.ufs.ac.za/biotech. Communicated by Carlien Pohl <pohlch@ufs.ac.za>.

James du Preez, former head of this department and a current commissioner of the International Commission on Yeasts, retired at the end of 2015. He remains with the department as a research fellow.

The following papers were recently published or accepted for publication:

1 Schabort DWP, Letebele PK, Steyn L, Kilian SG and du Preez J.C 2016 Differential RNA-seq, multi-network analysis and metabolic regulation analysis of *Kluyveromyces marxianus* reveals a compartmentalized response to xylose. PLoS ONE (accepted).

We investigated the transcriptomic response of a new strain of the yeast *Kluyveromyces marxianus*, in glucose and xylose media using RNA-seq. The data were explored in a number of innovative ways using a variety of networks types, pathway maps, enrichment statistics, reporter metabolites and a flux simulation model, revealing different aspects of the genome-scale response in an integrative systems biology manner. The importance of the subcellular localisation in the transcriptomic response is emphasised here, revealing new insights. As was previously reported by others using a rich medium, we show that peroxisomal fatty acid catabolism was dramatically up-regulated in a defined xylose mineral medium without fatty acids, along with mechanisms to activate fatty acids and transfer products of β-oxidation to the mitochondria. Notably, we observed a strong up-regulation of the 2-methylcitrate pathway, supporting capacity for odd-chain fatty acid catabolism. Next we asked which pathways would respond to the additional requirement for NADPH for xylose utilisation, and rationalised the unexpected results using simulations with Flux Balance Analysis. On a fundamental level, we investigated the contribution of the hierarchical and metabolic regulation levels to the regulation of metabolic fluxes. Metabolic regulation analysis suggested that genetic level regulation plays a major role in regulating metabolic fluxes in adaptation to xylose, even for the high capacity reactions, which is unexpected. In addition, isozyme switching may play an important role in re-routing of metabolic fluxes in subcellular compartments in *K. marxianus*.

2 Ogundeji AO, Albertyn J, Pohl CH and Sebolai OM 2016 Method for identification of *Cryptococcus neoformans* and *Cryptococcus gattii* useful in resource-limited settings. J Clin Pathol 69:352–357 - doi:10.1136/jclinpath-2014-202790

Aims The high HIV/AIDS burden in Sub-Saharan Africa has led to cryptococcosis becoming a public health concern. In this resource-limited setting, conventional identification methods are mainly used to diagnose cryptococcal infections. However, these methods are often inconsistent, and importantly, cannot discriminate between the aetiological agents, *Cryptococcus neoformans* and *C. gattii*. Therefore, there is a need for an alternative reliable method to identify these species. **Methods** We examined the usefulness of a PCR method, including a restriction digest, in identifying clinical *C. neoformans* and *C. gattii* isolates. In addition, matrix-assisted laser desorption ionisation time-of-flight mass spectrometry (MALDI-ToF MS) was performed for validation purposes. **Results** The intraspecific variation between tested strains allowed for their delineation into three traditional varieties of *C. neoformans*, that is, varietal forms: *neoformans*, *grubii* and *gattii*. Furthermore, we uncovered a restriction site (signature sequence: 5'-AATATT-3') that is present only in the distinct species *C. neoformans* (varietal forms *neoformans* and *grubii*), and is, importantly, absent in the distinct species *C. gattii* (*C. neoformans* var. *gattii*). Thus, we

were able to discriminate the distinct species by directly digesting the PCR amplicons using the endonuclease SspI. It was also possible to delineate some tested isolates as either *C. neoformans* or *C. gattii* using our MALDI-ToF MS data.

Conclusions The possibility of performing only a restriction digest makes the outlined method, similar to conventional techniques, economical and easy to optimise for routine use in resource-limited settings.

3 Akanni GB, du Preez JC, Steyn L and Kilian SG 2015 Protein enrichment of an *Opuntia ficus-indica* cladode hydrolysate by cultivation of *Candida utilis* and *Kluyveromyces marxianus*. J Sci Food Agric 95:1094-1102 - doi: 10.1002/jsfa.6985

4 Madu UL, Ogundeji AO, Mochochoko BM, Pohl CH, Albertyn J, Swart CW, Allwood JW, Southam AD, Dunn WB, May RC and Sebolai OM 2015 Cryptococcal 3-hydroxy fatty acids protect cells against amoebal phagocytosis. Frontiers Microbiol 6: article 1351 - doi: 10.3389/fmicb.2015.01351

5 Motaung TE, Albertyn J, Pohl CH and Kőhler G 2015 *Candida albicans* mutant contruction and characterization of selected virulence determinants. J Microbiol Methods 115:153-165 - doi: 10.1016/j.mimet2015.06.004

6 Pienaar GH, Einkamerer OB, Van der Merwe HJ, Hugo A and Fair MD 2015 The effect of an active live yeast product on the digestibility of finishing diets for lambs. Small Ruminant Res 123:1-8 - doi: 10.1016/j.smallrumres.2014.11.001

XV Leibniz Institute DSMZ-German Collection of Microorganisms and Cell Cultures, Inhoffenstraße 7B, 38124 Braunschweig, Germany – www.dsmz.de. Communicated by AM Yurkov <andrey.yurkov@dsmz.de>.

During the Yeast Taxonomy workshop held in Utrecht last April, yeast taxonomy was discussed genus by genus, considering the changing nomenclature and the use of the 'One Fungus = One name' rule. These discussions were continued at the Yeast Taxonomy workshop during the ISSY32 meeting in Perugia last September. As a follow up of these discussions, a total of five papers will be published in Studies in Mycology, in a volume entitled "Multigene phylogeny and reclassification of yeasts and related filamentous taxa in Basidiomycota" (2015. Volume 81), which was edited by Teun Boekhout and Feng-Yan Bai. Papers published in this volume report the results of multi-gene phylogenetic analyses of basidiomycetous yeasts and introduce taxonomic changes in the Tremellomycetes, the Pucciniomycotina and the Ustilaginomycotina. In agreement with decisions made during taxonomy workshops, several large polyphyletic yeast genera such as *Bullera*, *Cryptococcus*, *Pseudozyma*, *Rhodotorula*, *Sporobolomyces*, *Tilletiopsis* and some more were reduced to phylogenetic clades containing the respective type species. Despite ongoing debates regarding the selection of anamorphic or teleomorphic names for reclassified yeast genera in order to comply with the new nomenclature, an older name was always chosen in the present taxonomic revision. As a result, several older names originally introduced for teleomorphic genera were resurrected and used to accommodate asexual and sexual species of the same phylogeneic genus. In agreement with taxonomic priority, the following anamorphic names were proposed to be used in the future, namely *Rhodotorula* (over *Rhodosporidium*) and *Sporobolomyces* (over *Sporidiobolus*) and *Sterigmatomyces* (over *Agaricostilbum*). Full texts of the papers are freely available at Studies in Mycology:
http://www.sciencedirect.com/science/journal/01660616/81

Recently published papers.

1 Mittelbach M, Yurkov AM, Stoll R, Begerow D 2016 Inoculation order of nectar-borne yeasts opens a door for transient species and changes nectar rewarded to pollinators. Fungal Ecology: in press. DOI: 10.1016/j.funeco.2015.12.003

Nectar-borne yeast communities are species poor assemblages compring is a few specialized taxa (Saccharomycotina) and many transient species. Short flower lifetimes and harsh environmental conditions impose an enormous pressure on nectar-colonizers, which try to overcome these challenges through fast multiplication and osmotolerance. Since these traits are exclusively known for ascomycetes, the origin of multi-species communities is still poorly understood. We conducted field and laboratory experiments to analyze the competition between autochthonous pollinator-borne and transient yeast species in nectar. Subsequently we analyzed the impact of microbial growth on the environment. Our results endorse theories on priority effects and show that yeast incidences in natural flowers, cell densities in microcosms and the environmental impact strongly depend on the inoculation order of the respective yeast species. Transient species are more frequent in flowers visited only once, while specialists require several flower visits to establish common population structures most probably through tough inner-floral competition.

2 Yurkov AM, Röhl O, Pontes A, Carvalho C, Maldonado C, Sampaio JP. 2016. Local climatic conditions constrain soil yeast diversity patterns in Mediterranean forests, woodlands and scrub biome. FEMS yeast research 16: fov103.

Soil yeasts represent a poorly known fraction of the soil microbiome due to limited ecological surveys. Here, we provide the first comprehensive inventory of cultivable soil yeasts in a Mediterranean ecosystem, which is the leading biodiversity hotspot for vascular plants and vertebrates in Europe. We isolated and identified soil yeasts from forested sites of Serra da Arrábida Natural Park (Portugal), representing the *Mediterranean forests, woodlands and scrub biome*. Both cultivation experiments and the subsequent species richness estimations suggest the highest species richness values reported to date, resulting in a total of 57 and 80 yeast taxa, respectively. These values far exceed those reported for other forest soils in Europe. Furthermore, we assessed the response of yeast diversity to microclimatic environmental factors in biotopes composed of the same plant species but showing a gradual change from humid broadleaf forests to dry maquis. We observed that forest properties constrained by precipitation level had strong impact on yeast diversity and on community structure and lower precipitation resulted in an increased number of rare species and decreased evenness values. In conclusion, the structure of soil yeast communities mirrors the environmental factors that affect aboveground phytocenoses, aboveground biomass and plant projective cover.

3 Pontes A, Röhl O, Carvalho C, Maldonado C, Yurkov AM, Sampaio JP 2016 *Cystofilobasidium intermedium* sp. nov. and *Cystofilobasidium alribaticum f.a.* sp. nov. isolated from Mediterranean forest soils. International Journal of Systematic and Evolutionary Microbiology 66: 1058-1062.

Multiple isolates belonging to the basidiomycetous genus Cystofilobasidium were obtained from forest soils in Serra da Arrábida Natural Park in Portugal. Phylogenetic analyses employing concatenated sequences of the D1/D2 domain and ITS region support the recognition of two novel species: *Cystofilobasidium alribaticum f.a.*, sp. nov. (type strain CBS 14164^T = PYCC 6956^T = DSM 101473^T) and *Cystofilobasidium intermedium* sp. nov. (type strain CBS 14089^T = PYCC 6856^T = DSM 101474^T). Whereas *C. alribaticum f.a.* sp. nov. does not form hyphae, even when different strains are crossed, *C. intermedium* sp. nov. is self-fertile and forms mycelium with teliospores that upon germination give rise to slender basidia. The most remarkable physiological trait of the two novel species is their ability to grow at 35 °C, a property not observed for remaining species of the genus.

4 Boundy Mills KL, Glantschnig E, Roberts IN, Yurkov A, Casaregola S, Daniel HM, Groenewald M, Turchetti B 2016 Yeast culture collections in the twenty first century: New opportunities and challenges. Yeast: in press. DOI: 10.1002/yea.3171

5 Liu XZ, Wang QM, Göker M, Groenewald M, Kachalkin AV, Lumbsch HT, Millanes AM, Mats Wedin M, Yurkov AM, Boekhout T, Bai FY 2015 Towards an integrated phylogenetic classification of the Tremellomycetes. Studies in Mycology 81: 85-147.

6 Wang QM, Yurkov AM Göker M, Lumbsch HT, Leavitt SD, Groenewald M, Theelen B, Liu XZ, Boekhout T, Bai FY 2015 Phylogenetic classification of yeasts and related taxa within Pucciniomycotina. Studies in Mycology 81: 149-189.

XVI Department of Biology, University of Western Ontario, London, Ontario, Canada N6A 5B7. Communicated by MA Lachance <lachance@uwo.ca>.

Recent publications.

1 Possmayer M, Gupta RK, Szyszka-Mroz B, Maxwell DP, Lachance MA, Hüner NPA, Smith DR 2015 Resolving the phylogenetic relationship between *Chlamydomonas* sp UWO241 and *Chlamydomonas raudensis* SAG 49.72 (Chlorophyceae) with nuclear and plastid DNA sequences. J Phycol 52:305–310.

2 Lachance MA, Hurtado E, Hsiang T 2016 A stable phylogeny of the large-spored *Metschnikowia* clade. Yeast (in press) - DOI: 10.1002/yea.3163.

Draft genomes of 55 strains representing all known large-spored *Metschnikowia* species were used to construct a robust phylogeny of these yeasts found in association with flower-visiting insects. The genomes were annotated with reference to *Clavispora lusitaniae*. From 3016 orthologues identified, 1061 were present in all strains with enough overlap to generate alignments of 500 bp or more. We constructed trees for all those alignments and evaluated their accuracy from their ability to resolve each of 22 sets of conspecifics correctly as sister taxa. Neighbour-joining identified species membership better than maximum likelihood, as did trees based on larger gene alignments. However, correct species assignment was not predictive of a gene's ability to resolve deeper topologies, which were more reliably identified by maximum likelihood analyses of large concatenations. Specifically, 14 trees based on independent concatenations ca. 100 kb in length were topologically consistent with a tree based on a single, large concatenation (1,410,065 positions), lending a high degree of confidence to the stability of the phylogeny. A tree based on a concatenation of intergenic regions (112 136 positions) was also congruent. Again, the best predictor of phylogenetic signal quality of a gene was the size of the alignment. Bootstraps were not always good indicators of phylogenetic quality, as they were sometimes affected by clade size. A tree constructed from a presence-absence matrix of all annotated genes was remarkably congruent with sequence-based phylogenies, suggesting that gain or loss of genes is worth exploring further as a phylogenetically significant event.

3 Lopes M, Morais C, Kominek J, Cadete R, Soares MA, Uetanabaro AP, Fonseca C, Lachance MA, Hittinger C, Rosa CA 2016 Genomic analysis and D-xylose fermentation of three novel *Spathaspora* species: *Spathaspora girioi* sp. nov., *Spathaspora hagerdaliae* f. a., sp. nov., and *Spathaspora gorwiae* f. a., sp. nov. FEMS Yeast Res (in press) - doi.org/10.1093/femsyr/fow044

See abstract under Dr. Hittinger's communication.

Half a Century of the International Commission on Yeasts

As indicated in the first page, the Yeast Newsletter is the official publication of the International Commission for Yeasts (ICY) of the International Union of Microbiological Societies (IUMS). Many of the young readers of the Newsletter may not know what ICY is and what is its history. ICY is an active international body, whose role is "to establish effective liaison between persons and organizations concerned in yeast investigations, and between them and the practical users of results of investigations, including yeast culture collections". This year we commemorate the fiftieth anniversary of its existence. When and how did the story of ICY start? The creation of this body is linked to the second International Symposium on Yeasts which was held in Bratislava, Czechoslovakia, now the capital of Slovakia, in July 1966. The meeting was attended by 145 participants from 21 countries. During the Symposium the Czechoslovak representatives initiated the creation of an international organization which would stimulate scientific collaboration of people working with yeasts all over the world. A Council for International Collaboration in Yeast Science was founded. The late Dr. A. Kockova-Kratochvilova (1915-1992) was appointed Chair and Dr. Erich Minarik (1924-2007), Secretary of the Council; both were from Czechoslovakia. The other endowing members of the Council were: K. Beran (Czechoslovakia), A. Eddy (UK), P. Elinov (USSR), H. Klaushoffer (Austria), N.I. Kudrjavcev (USSR), U. Leopold (Switzerland), R. Muller (GDR), S. Nagai (Japan), O. Necas (Czechoslovakia), H.J. Phaff (USA), C.F. Robinow (Canada), H. Soumalainen (Finland), T. Tsuchiya (Japan), L. Wickerham (USA), T. Wikén (Holland), and S. Windisch (GFR), all well-recognized yeast researchers at the time. During the meeting it was also agreed that the existing Yeast Newsletter, edited by H.J. Phaff at the University of California, would serve as the official publication of the Council. Thanks to Prof. Phaff and later contributors, the Yeast Newsletter is still alive and bringing interesting and important information to our groups.

In early years after its foundation, the Council underwent changes in names and affiliations. In 1970, the International Commission on Yeasts and Yeast-like Microorganisms (ICY) became a part of the Microbiology Division of the International Union of Biological Sciences (IUBS) and in 1981, the Mycology Division of the International Union of Microbiological Societies.

The main activity of ICY is the organization of the International Congress on Yeasts at 4 years intervals (until 2004 International Symposium on Yeasts) and nearly annually, the International Specialized Symposium on Yeasts (ISSY). The conferences are held in different countries. The main organizer of the International Congress becomes Chair of the ICY for a four-year term. The current Chair is Dr. Charles A. Abbas from Archer Daniels Midland Company (IL, USA) who was co-organizer of the successful 13th ICY in Madison, WI, USA, in August 2012. He will hold the Chair till the 14th ICY, which is being organized by Prof. Hiroshi Takagi and will be held on Awaji Island in Hyogo Prefecture, Japan in September 2016.

On behalf of the readers and the Editorial Board of the Newsletter, let us wish the International Commission on Yeasts a long future of successful activities and achievements.

Peter Biely, Associate Editor
Marc-André Lachance, Editor

Forthcoming Meetings

14th International Congress on Yeasts (ICY14)
Awaji Yumebutai International Conference Center
Awaji Island, Japan, September 11-15, 2016

On behalf of the Organization Committee, I'm great honored to host the 14th International Congress on Yeasts (ICY14) at Awaji Yumebutai International Conference Center, Awaji Island, Japan, in September 11-15, 2016.

The general topic of ICY14 is "Yeasts for Global Happiness". It means that yeast science & technology will contribute to the world in terms of food & beverage, health & medicine, energy & environment. In addition, this congress will be the first time held in Japan since ISSY2 in back to 1972, when I was a junior high school student. We'd like to send valuable message and information from Japan to the world. More importantly, I believe that this will be a great opportunity for young scientists as they can attend and present their research. In contrast, we senior must tell the fun and importance of yeast research to the next generation.

ICY14 is sponsored by the International Commission on Yeasts as part of the International Union of Microbiological Societies (IUMS). The ICY has been held once every four years since 1955. It provides an opportunity for presenting the latest research progress in yeast metabolism, physiology, genetics, genomics, regulation, ecology, systematics, phylogeny, food and beverage applications, biofuel production and clinical applications.

I am very much looking forward to seeing you on middle September in 2016 at Awaji Island, Japan!

Hiroshi Takagi, Head of the Organizing Committee of ICY14 http://icy2016.com/index.html

Workshop: The Yeasts, a Taxonomic Study
ICY 2016 - September 12 2016

Since the publication of the fifth edition of The Yeasts, a taxonomic Study in 2011 many things related to yeast taxonomy have changed. Concepts of several genera changed, and many new species have been described. Moreover, the 'One Fungus = One Name' principle has started to be implemented.

During several yeast meetings that were held during the last years the future of 'The Yeasts' has been discussed. In May 2016, the editors for the future edition had a discussion meeting and we would like to present and discuss the outcome, which is largely based on recommendations made during the various discussions held in the past, during the forthcoming ICY14 in Japan.

On September 12 there will be a lunch discussion followed by an afternoon workshop session. During these meetings we hope to reach maximum consensus to further develop 'The Yeasts' in the future. We hope to see many of you in Japan.

On behalf of all editors,
Teun Boekhout <t.boekhout@cbs.knaw.nl>

Yeasts as Versatile Testbeds for the Life Sciences Madrid, Spain, in October 17th-18th 2016

The Fundación Ramón Areces sponsors in Madrid (Spain) in October 17th-18th 2016 a Symposium with the title "Yeasts as versatile testbeds for the life sciences" coordinated by Carlos Gancedo (Madrid) and Jack Pronk (Delft). The speakers and the topics are the following:

Alcalde, M. *Saccharomyces cerevisiae* as a tool-box for protein engineering by directed evolution; **Branduardi, P.** Yeasts and stress responses: learning how to leverage cellular potential for matching industrial requirements; **Daran, J.M.** CRISPR/Cas9: a molecular Swiss army knife. From gene to pathway to genome engineering; **Gancedo, C.** The rise of moonlighting proteins in yeasts, **Gil, C.** Candida albicans-macrophage interaction: insights from proteomics; **González, R.** Yeast physiological diversity and interspecies interactions under industrially relevant conditions; **Holthuis, J.** Yeast-based cloning and functional analysis of a candidate ceramide sensor from mammals; **Kaeberlein, M.** From mTOR to mitochondria: How aging yeast are providing mechanistic insights in translational geroscience, **Liti, G.** Yeast population genomics: origin and evolution of a classic model organism; **Machín, F.** Budding yeast as a model system to study the causes and consequences of anaphase bridges; **Perez-Ortin, J.** Degradation/synthesis cross talk during mRNA turnover; **Posas, F.** Understanding control of cell proliferation from yeast to mammals; **San-Segundo, P.** Why does (sometimes) meiosis fail? Lessons from yeast to prevent gamete aneuploidy, **Smolke, C.** To be announced; **Winderickx, J.** Protein folding diseases: Lessons learned from yeast; **Zamboni, N.** Reconstructing metabolism by high-throughput mass spectrometry.

Carlos Gancedo <cgancedo@iib.uam.es>

9th International Fission Yeast Meeting, May 14-19, 2017, in Banff, Alberta, Canada

The 9th International Fission Yeast Meeting is coming to Canada and will be held May 14-19, 2017, in Banff, Alberta, at the Banff Centre Conference Facility. This is particularly exciting because the first Fission Yeast Workshop was held in conjunction with the Thirteenth International Congress on Yeast Genetics and Molecular Biology at the Banff Centre in 1986. In 2017 we are expecting 350-450 guests representing Pombe labs from around the globe. The organizing committee is comprised of Paul Young, Queen's, Gordon Chua, Calgary, and Dallan Young, Calgary. For further information, contact:

Paul G. Young
Biology, 2443 Biosciences,
Queen's University,
Kingston, ON K7L 3N6 <youngpg@queensu.ca>

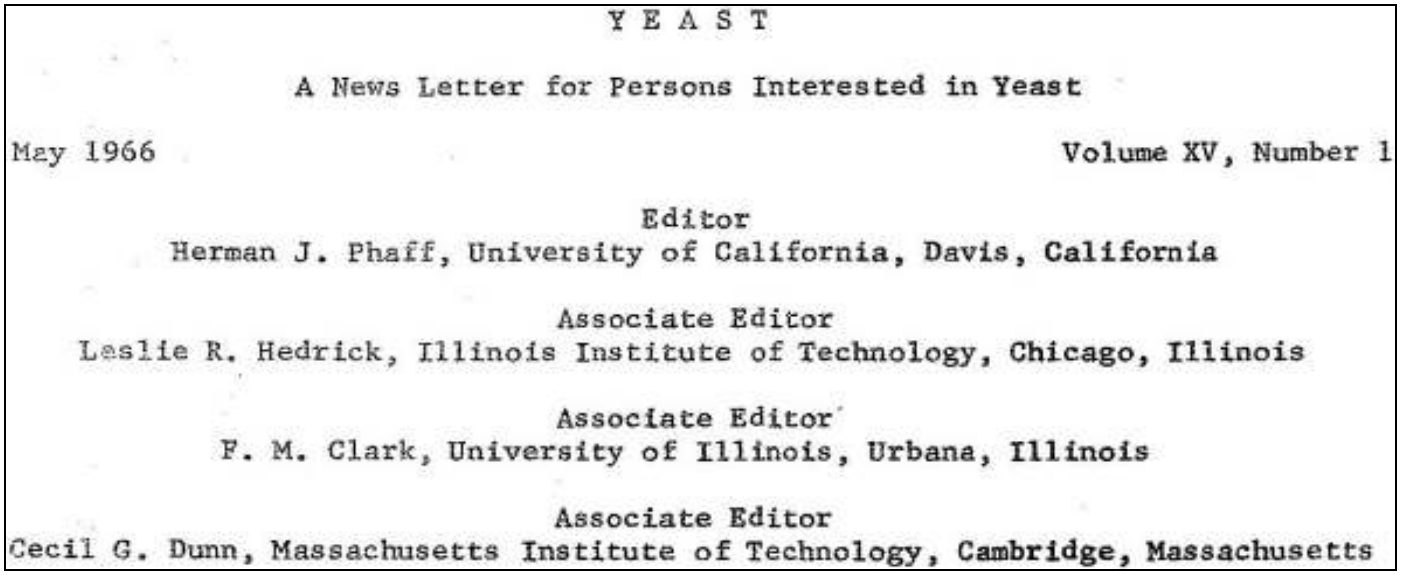
Brief News Item

Postdoctoral/Researcher Position Sought

I have defended my PhD thesis in Molecular Biology entitled: "The development of a new expression system based on the new species of methylotrophic yeast *Komagataella kurtzmanii*". The work was done under the supervision of Dr. Dmitry Kozlov, head of the laboratory of eukaryotic expression systems, GosNIIgenetika, Moscow, Russia - www.genetika.ru. I have gained experience with the optimization of heterologous expression in yeast (*P. pastoris*, *K. kurtzmanii*, *S. cerevisiae*, *K. lactis*) as well as with metabolic engineering in *S. cerevisiae*. I currently work also at the Ajinomoto Genetics Research Institute as a research associate. I am looking for a postdoctoral or researcher position. A summary of my research can be found at https://www.researchgate.net/profile/Oleg_Tyurin2

Oleg Tyurin <oleg.tyurin77@gmail.com>

50 Years Ago



Y E A S T

A News Letter for Persons Interested in Yeast

May 1966 Volume XV, Number 1

Editor
Herman J. Phaff, University of California, Davis, California

Associate Editor
Leslie R. Hedrick, Illinois Institute of Technology, Chicago, Illinois

Associate Editor
F. M. Clark, University of Illinois, Urbana, Illinois

Associate Editor
Cecil G. Dunn, Massachusetts Institute of Technology, Cambridge, Massachusetts

Miss W. Ch. Slooff (CBS, the Netherlands) reported that type strains of 21 new yeast species were deposited in the collection. [Note: This was a major increase from 7 species reported in fall 1965.]

Dr. James A. Barnett (Low Temperature Research Station, Cambridge, England) described a method to scrutinize the results of taxonomic growth tests on different carbon sources, based on analyses of *Endomycopsis, Pichia* and *Debaryomyces* done by Kreger-van Rij, and *Candida* and *TorulopsisTorulopsis* done by van Uden. He requested collaboration with other researchers who analyzed large groups of strains on different substrates, suggesting discussion at the International Yeast Symposium to be held in Bratislava.

Yeasts on grapes, in grape juice and wines in Czechoslovakia were systematically studied by **Dr. E. Minarik** (Bratislava, Czechoslovakia) They found that *Kloeckera apiculata, Candida pulcherrima* and *Saccharomyces cerevisiae* var. *ellipsoideus* were dominant in the early stages of spontaneously fermenting juice, and *Saccharomyces oviformis* dominated in the final stages of fermentation. No direct influence of soil conditions or grape variety could be established.

Professor Samuel P. Meyers (University of Miami, Florida, USA) anticipated participating in the August 1966 meeting of the U.S. –Japan Seminar in Marine Microbiology to be held in Tokyo, Japan, and listed four recent publications on marine yeasts.

Professor J. B. Sinclair (Louisiana State University, USA) shared the abstract of a recently published paper on plant and animal isolates of *Geotrichum candidum*.

Dr. H. Jean Shadomy (Medical College of Virginia, USA) requested additional strains of *Cryptococcus neoformans* to add to the 43 strains gathered for their ongoing studies of hyphal formation.

Dr. Shoji Goto (Yamanashi University, Kofu, Japan) shared the abstract of a publication in press regarding yeasts isolated from oil brines from oilfields in Japan.

Dr. Colin Clarke (Institute of Animal Genetics, Edinburgh, Scotland) listed two recently published papers on UV-induced adenine mutants of *Schizosaccharomyces pombe*.

Dr. H. Gutz (Graduate Research Center of the Southwest, Dallas, Texas, USA) found that diploid strains of *Schizosaccharomyces pombe* could be haploidized by treatment with p-fluorophenylalanine. They hoped to establish, with haploidization experiments, the number of chromosomes in this yeast.

Professor C. C. Lindegren (Southern Illinois University, Illinois, USA) reported publication of three articles related to yeast mitochondria, food yeast and the lysine biosynthetic pathway.

Dr. J. G. Kleyn (University of Puget Sound, Tacoma, Washington, USA) shared the abstract of a paper on formation of dwarf cells by yeasts other than *Saccharomyces carlsbergensis*, presented at the American Association for the Advancement of Science (AAAS) annual meeting.

Professor Dr. S. Windisch (Institut für Gärungsgewerbe, Berlin, West Germany) described studies on the microbiology of Marzipan, analysis of bottom fermenting *Saccharomyces* yeasts, and methods to detect *Saccharomyces rouxii* in high-sugar products.

Dr. H. J. Phaff (University of California Davis, USA) described work being done by predoctoral student Mr. Edward Buecher on in vitro growth of *Saccharomycopsis guttulata,* in which they discovered a need for higher CO_2 concentrations. Strains of this yeast isolated from wild black-tailed jack rabbits differed in morphology from strains isolated from domestic rabbits. Phaff also presented the abstract of a paper to be presented at the International Yeast Symposium at Bratislava, on exo-β-glucanase in yeast.

Miss F. R. Elliott and Mr. M. Richards (The Brewing Industry Research Foundation, Surrey, England) summarized a publication in Nature regarding selective isolation of yeasts from a contaminated environment using media containing streptomycin, and another publication on use of "giant colonies" to distinguish between brewing strains of *Saccharomyces cerevisiae*.

Dr. J. O. Lampen (Rutgers, New Brunswick, New Jersey, USA) presented a review of the action of the polyenic antifungal antibiotics at the April 1966 symposium of the Society for General Microbiology. They also shared the abstract of a presentation by Dr. Norbert Neumann regarding properties of a non-repressible yeast invertase produced by a yeast mutant obtained by UV irradiation by Miss Bland Symington.

Professor F. M. Clark (University of Illinois, USA) described the purification and analysis of the extracellular polysaccharide produced by *Torulopsis melibiosum*. It appears similar to the extracellular polysaccharide produced by *Lipomyces starkeyi*, which was reported in the previous Yeast News Letter.

Dr. M. E. Slodki (U.S. Department of Agriculture, Peoria, Illinois, USA) reported an abstract of a recently published paper in the Journal of General Microbiology on extracelluar polysaccharides and classification of *Lipomyces*, co-authored by L. J. Wickerham.

Dr. D. Pappagianis (University of California Berkeley, USA) detected very low levels of ethanol produced by several strains of *Cryptococcus neoformans* when grown in glucose-containing medium. Detection of ethanol in spinal fluid as a means of diagnosis of cryptococcal meningitis therefor appears to be of limited diagnostic value.

Dr. Cavit Akin (Falstaff Brewing Corporation, St. Louis, Missouri, USA) presented three papers at the American Society of Brewing Chemists Convention in Toronto, Canada on yeast sedimentation in beer fermentation, deep fermentation, and fermentation technology.

Brief News Items:

Dr. M. Ingram of the Low Temperature Research Station, Cambridge, England reported that the station would be closed in the end of 1967, and programs moved to two new facilities. J. A. Barnett will go to the Food Research Institute near Norwich, and M Ingram wil go to the Meat Research Institute near Bristol.

Dr. J. W. Fell recently completed a 3 month cruise in the Antarctic to study yeasts in ocean waters. He will begin a year of postdoctoral work in the laboratory of Dr. H. J. Phaff starting in August 1966.

Dr. Wm. Bridge Cooke (Department of Health, Education, and Welfare, Cincinnati, Ohio, USA) announced that laboratory work on isolation and identification of yeasts from sewage and polluted waters has been discontinued. The 5,000 yeast strains examined were not retained, but data sheets are being used to develop a comprehensive summary.

Kyria Boundy-Mills, Phaff Yeast Culture Collection, University of California Davis

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