

A Newsletter for Persons Interested in Yeast

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Editorials

Prof. Martin W. Miller 1925-2005

I have learned with great sadness the recent passing of Dr. Martin Miller, at the age of 80. To many of us, Marty was our beloved friend and colleague as well as our respected professor at the University of California, Davis. He will be missed.

Change in Editorial Board

Dr. Yasuji Oshima, of Kansai University in Osaka, recently retired. For several years, Dr. Oshima served on the Editorial Board of the Yeast Newsletter, assisting by accepting subscriptions from Japanese readers. On behalf of the Board and all readers, I thank him wholeheartedly for his efforts and wish him a happy retirement.

Printed and Electronic Subscriptions

A reminder of some recent changes in the subscription rates and modalities. The **printed version** of the Yeast Newsletter will continue to be available to readers for USD\$8.00 (Canada and U.S.A.) or USD\$12.00 (all other countries). The subscription is due upon receipt of the invoice that accompanies the December issue. Credit card payments can only be accepted for payments of USD\$40.00 or more.

The **electronic version** is sent free of charge to readers whose accounts are in order. To be added to the electronic mailing list, please email me at <u>lachance@uwo.ca</u>.

Readers who had not renewed for 2005 were sent, in April, a reminder card indicating the amount in arrears (if any). Please note that readers who did not reply to that reminder card will be removed from the mailing list.

Yeast Newsletter / International Commission on Yeasts - Website

http://publish.uwo.ca/~lachance/YeastNewsletter.html

Readers are encouraged to browse the YNL website and make suggestions for additions or improvements. Currently, the website contains a listing of positions available, lists of past and future conferences, and many YNL back issues. Readers are invited to send URLs of relevant web sites, including their own, to be added as links to the YNL home page. Please be sure to bookmark the YNL website and also to add a link to the YNL in your own webpage.

Back Issues

We are still missing issues of the YNL published prior to November 1958 and would welcome these.

M. A. Lachance Editor

I. Department of Food Science and Technology, Dalhousie Sexton Campus, P.O. Box 10001360 Barrington St, Halifax, Nova Scotia, Canada. Communicated by A. Speers - <u>Alex.Speers@dal.ca</u>

Research in progress.

1. C Jenkins, DA Wood, T Gill & A Speers. Impact of malted barley quality and wort composition on the occurrence of premature yeast flocculation

Premature yeast flocculation (PYF) is characterized by aberrant fermentation profiles. Malt constituents are believed to cause of PYF. Worts, (from control & PYF malt) were fermented and analysed for wort constituents yeast growth, attenuation and cell surface physical properties (hydrophobicity, zeta potential, Helm's flocculation, orthokinectic capture coefficient, zymolectin and mannose receptor densities). Worts exhibited differences in peptide, carbohydrate, FAN and metal ion composition. PYF wort exhibited aberrant fermentation profiles after 60 h when modeled by a logistic equation. As little effect on cell properties were noted it is proposed that two PYF behaviors: PYF-M (metabolic) and PYF-B (binding) exist.

II. Dipartimento di Scienze e Tecnologie Agro-Forestali e Ambientali, Università di Reggio Calabria, Piazza San Francesco 7, I-89061 Gallina (RC), Italia. Communicated by A. Caridi acaridi@unirc.it

Recent publications.

1. Caridi A, Pulvirenti A, Restuccia C & Sidari R 2005 Screening for yeasts able to hydrolyse arbutin in the presence of glucose or ethanol. Annals Microbiol **55**:43-46.

The study was carried out using 212 glucose-fermenting yeasts isolated from Calabrian and Sicilian samples of must and wine. They were screened for β -glucosidase activity in Petri plates containing arbutin agar medium. Eleven strains (three apiculate yeasts and eight elliptic yeasts) were able to perform arbutin hydrolysis, but only strain C5, identified as *Pichia*

anomala, maintained the ability to hydrolyse arbutin even in the presence of remarkable amounts of glucose or ethanol. This strain did not show the capacity to excrete the β -glucosidase into the medium; effectively, the enzymatic activity in the culture supernatant was near to zero. However, its intracellular activity was high.

2. Caridi A. Enological functions of the parietal yeast mannoproteins. First International Conference on Environmental, Industrial and Applied Microbiology, BioMicroWorld2005, Badajoz (Spain), 15-18 March 2005.

Mannoprotein production and release, both during winemaking and during aging on lees, depends on the specific yeast strain, as well as the nutritional conditions. The following enological functions of the parietal yeast mannoproteins have been described: a) improvement of the growth of malolactic bacteria; b) prevention of potassium bitartrate precipitation; c) prevention of proteic haze in white wines; d) interactions with aroma compounds; e) interactions during barrel aging on fine lees; f) interactions with sparkling wines; g) interactions with flor sherry-type products; h) interactions with phenolic compounds; i) adsorption of ochratoxin A. It appears that parietal mannoproteins can play a very important role in the overall vinification process. Further discoveries related to their enological functions are foreseeable. Yeast-derived mannoproteins may well induce chemical, sensorial and health benefits, thus greatly improving wine quality.

3. Caridi A., Galvano F., Tafuri A., Ritieni A. Ochratoxin A removal during alcoholic fermentation. First International Conference on Environmental, Industrial and Applied Microbiology, BioMicroWorld2005, Badajoz (Spain), 15-18 March 2005.

This work aims to investigate the performance of twenty strains of *Saccharomyces sensu stricto* to remove ochratoxin A (OTA) during winemaking. Each strain was inoculated in triplicate in 10 mL of white must naturally contaminated with OTA (1.58 ng/mL), and again in the same must artificially contaminated with OTA (7.63 ng/mL). This microvinification trial was performed at 25°C for 90 days. The toxin content and the ethanol content of the wines were assayed; the toxin content of the lees was also analysed separately. The OTA content in wines produced from the naturally contaminated

must varies from 0.143 to 0.950 ng/mL (mean 0.498 ng/mL). The OTA content in wines obtained from the artificially contaminated must varies from 1.270 to 2.448 ng/mL (mean 1.661 ng/mL). The OTA content in lees varies from 1.537 to 7.456 ng/mg of biomass (wet weight). These last extremely interesting results indicate that OTA-removal from grape must was probably carried out by the yeast cell wall, acting like a sponge. A role of the parietal yeast mannoproteins in OTA adsorption was hypothesised and the implications of these results in the winemaking of OTA-contaminated musts were discussed.

III. State Scientific-Research Institute for Genetics and Selection of Industrial Microorganisms, I-Dorozhnyi 1, Moscow 117545, Russia. Communicated by G.I. Naumov and E.S. Naumova gnaumov@yahoo.com

Many thanks to B. Dujon (Paris), S. Casaregola (Grignon, France), M.-A. Lachance (London, Canada), V. Larionov (Bethesda, USA), E. Kroll (Berkeley, USA) and A.J.S. Klar (Frederick, USA) for the opportunity to visit their labs.

The following are publications for 2004-2005 or in press

- 1. Naumova E.S., Sukhotina N.N., Naumov G.I. 2004. Molecular-genetic differentiation of the dairy yeast *Kluyveromyces lactis* and its closest wild relatives. FEMS Yeast Res., 5: 263-269.
- 2. Naumova E.S., Gazdiev D.O., Naumov G.I. 2004. Molecular divergence of the soil yeasts *Williopsis* sensu stricto. Microbiology (Moscow), 73 (6): 658–665.
- 3. NaumovG.I., Ivannikova Yu.V, Naumova E.S. 2005. Molecular polymorphism of viral dsRNA of yeast *Saccharomyces paradoxus*. Molecular genetics, microbiology and virology (Moscow), 1: 38-40.
- 4. Naumova E.S., Zholudeva M.V., Martynenko N.N., Naumov G.I. 2005. The molecular-genetic differentiation of cultured *Saccharomyces* strains. Microbiology (Moscow), 74 (2): 179-187.
- 5. Naumov G.I. 2005. Domestication of dairy yeasts *Kluyveromyces lactis*: transfer of the of β -galactosidase (*LAC*4) and lactose permease (*LAC*12) gene cluster? Dokl. Biol. Sci., 401: 120-122.
- 6. Sukhotina N.N., Naumova E.S., Naumov G.I. 2005. Comparative molecular-genetic analysis of clinical *Kluyveromyces lactis* strains. Advances in Medical Mycology. Proceedings of Third Congress on Medical Mycology. Moscow: All-Russian National Academy of Mycology, Vol. 5: 20-21 (in Russian).
- 7. Korshunova I.V, Naumova E.S., Naumov G.I. 2005. Comparative molecular-genetic analysis of the âfructosidases in yeast *Saccharomyces*. Molecular Biology (Moscow) (in press).

To infer the molecular evolution of polymeric β -fructosidase *SUC* genes of the yeast *Saccharomyces*, we have cloned and sequenced a new *SUC* gene from *S. cariocanus* and determined the sequence similarity of \hat{a} -fructosidases within the genus *Saccharomyces*. The proteins of *Saccharomyces cerevisiae* and its five sibling species (*S. bayanus*, *S. cariocanus*, *S. kudriavzevii*, *S. mikatae*, *S. paradoxus*) have high degree of identity – 90-97%. The invertase of *S. bayanus* is the most divergent among the proteins studied. The data obtained

indicated that the yeast invertases are highly conservative. In the coding regions of the *SUC* genes the pyrimidine transitions were the most abundant event due to silent changes mainly in the third codon position. There is only one, probably, non-telomeric *SUC* gene in each of the *Saccharomyces* species. In *S. cerevisiae*, *S. bayanus*, *S. kudriavzevii*, *S. mikatae* and *S. paradoxus* the *SUC* gene have been mapped on chromosome IX, whereas in *S. cariocanus* this gene is located in chromosone XV, in the position of translocation.

8. Naumova E.S., Ivannikova Yu.V., Naumov G.I. 2005. Genetic differentiation of sherry yeasts *Saccharomyces cerevisiae*. Appl Biochem Microbiol (Moscow) (in press).

Using different molecular methods we compared *S.cerevisiae* strains isolated at different stages of sherry process (young wine, criadera, solera) in different wine-making regions of Spain. The sherry yeasts were found to be divergent from the yeasts of primary wine fermentation on many physiological and molecular markers. All sherry strains, independently of their

origin, are characterised by the presence of the 24-bp deletion in the ITS1 of rDNA region. This deletion is absent in strains isolated from primary wine fermentation. Comparative study of the chromosomal DNAs indicates a similarity of molecular karyotypes of sherry strains from different populations.

IV. Department of Cell and Organism Biology, Söölvegatan 35, Lund University, SE 22362 Lund, Sweden. Communicated by J. Piškur - jure.piskur@cob.lu.se

Recent publications

1. J Piškur & RB Langkjær 2004. Yeast genome sequencing: the power of comparative genomics. Mol Microbiol 53:381-389.

For decades, unicellular yeasts have been general models to help understand the eukaryotic cell and also our own biology. Recently, over a dozen yeast genomes have been sequenced, providing the basis to resolve several complex biological questions. Analysis of the novel sequence data has shown that the minimum number of genes from each species that need to be compared to produce a reliable phylogeny is about 20. Yeast has also become an attractive model to study speciation in eukaryotes, especially to understand molecular mechanisms behind the establishment of reproductive isolation. Comparison of closely related species helps in gene annotation and to answer how many genes there really are within the genomes. Analysis of non-coding regions among closely related species has provided an example of how to determine novel gene regulatory sequences, which were previously difficult to analyse because they are short and degenerate and occupy different positions. Comparative genomics helps to understand the origin of yeasts and points out crucial molecular events in yeast evolutionary history, such as whole-genome duplication and horizontal gene transfer(s). In addition, the accumulating sequence data provide the background to use more yeast species in model studies, to combat pathogens and for efficient manipulation of industrial strains.

2. M Mentel, J Piškur, C Neuvéglise, A Ryčovská, G Cellengová, and J Kolarov 2005 Triplicate genes for mitochondrial ADP/ATP carriers in the aerobic yeast *Yarrowia lipolytica* are regulated differentially in the absence of oxygen. Mol Gen Genomics 273:84–91.

Yarrowia lipolytica is a strictly aerobic fungus, which differs from the extensively studied model yeasts *Saccharomyces cerevisiae* and *Schizosaccharomyces pombe* with respect to its physiology, genetics and dimorphic growth habit. We isolated and sequenced cDNA and genomic clones (*YIAAC1*) from *Y. lipolytica* that encode a mitochondrial ADP/ATP carrier. The *YI-AAC1* gene can complement the *S. cerevisiae* Daac2 deletion mutant. Southern hybridization, analysis of *Yarrowia* clones obtained in the course of the Génolevures project, and further sequencing revealed the existence of two paralogs of the *YIAAC1* gene, which were named *YIAAC2* and *YIAAC3*, respectively. Phylogenetic analysis showed that *YIAAC1* and *YIAAC2* were more closely related to each other than to *YIAAC3*, and are likely

to represent the products of a recent gene duplication. All three *Y. lipolytica YlAAC* genes group together on the phylogenetic tree, suggesting that *YlAAC3* is derived from a more ancient duplication within the *Y. lipolytica* lineage. A similar branching pattern for the three *ScAAC* paralogs in the facultative anaerobe *S. cerevisiae* demonstrates that two rounds of duplication of *AAC* genes occurred independently at least twice in the evolution of hemiascomycetous yeasts. Surprisingly, in both the aerobic *Y. lipolytica* and the facultative anaerobe *S. cerevisiae*, the three paralogs are differentially regulated in the absence of oxygen. Apparently, *Y. lipolytica* can sense hypoxia and down-regulate target genes in response.

V. Department of Applied Microbiology and Food Science, University of Saskatchewan, 51 Campus Drive, Saskatoon, Canada S7N 5A8. Communicated by W.M. (Mike) Ingledew ingledew@sask.usask.ca

Dr. E. Bellissimi has moved to Delft, Holland, to work at the Delft University of Technology as a postdoctoral fellow. Mr. Derek Abbott has joined the Delft University of Technology as a PhD student. We wish these two excellent students well in their further studies at this renowned institution.

The following papers have been published since our last report.

1. Bayrock D and WM Ingledew 2004 Inhibition of yeast by lactic acid bacteria in continuous culture: nutrient depletion and/or acid toxicity? J Ind Microbiol Biotechnol **31**:362-368.

Lactic acid was added to batch very high gravity (VHG) fermentations, and to continuous VHG fermentations equilibrated to steady state with *Saccharomyces cerevisiae*. A 53% reduction in CFU/ml of *S. cerevisiae* was observed in continuous fermentation at an undissociated lactic acid concentration of 3.44% w/v, and greater than 99.9% reduction was evident at 5.35% w/v lactic acid. The differences in yeast cell number in these fermentations were not due to pH since batch fermentations over a pH range of 2.5 to 5 did not lead to changes in growth rate. Similar fermentations performed in batch showed that growth inhibition with added lactic acid was nearly identical. This indicates that the apparent high resistance of *S. cerevisiae* to

lactic acid in continuous VHG fermentations is not a function of culture mode. Although the total amount of ethanol decreased from 48.7 to 14.5 g/L when 4.74% w/v undissociated lactic acid was added, the specific ethanol productivity increased ~3.2 fold (from 7.42 x 10^{-7} to 24.0 x 10^{-7} g ethanol per CFU per h) which indicated that lactic acid stress improved ethanol production of each surviving cell. In multistage continuous fermentations, lactic acid was not responsible for the 83% CFU/ml reduction of viable *S. cerevisiae* yeasts when *Lactobacillus paracasei* was introduced to the system at a controlled pH of 6. The competition for trace nutrients in those fermentations and not lactic acid produced by *L. paracasei* likely resulted in yeast inhibition.

2. DA Abbott and WM Ingledew 2004 Buffering capacity of whole corn mash alters concentrations of organic acids required to inhibit growth of *Saccharomyces cerevisiae* and ethanol production. Biotechnol Lett **26**:1313-1316.

Growth of *Saccharomyces cerevisiae* and fermentative ethanol production in the presence of acetic and lactic acids was measured in whole corn mash. In this industrial medium, as compared to glucose minimal medium, *Saccharomyces* showed increased tolerance to organic acid stress. It was concluded that the increased buffering capacity of whole corn mash, resulting in decreased concentration of undissociated acid, was responsible for this phenomenon.

3. DP Bayrock and WM Ingledew 2005 Ethanol production in multistage continuous, single stage continuous, Lactobacillus-contaminated continuous, and batch fermentations. World J Microbiol Biotechnol **21**:83-88.

Saccharomyces cerevisiae-based ethanol fermentations were conducted in batch culture, in a single stage continuous stirred tank reactor (CSTR), a multistage CSTR, and in a fermentor contaminated with *Lactobacillus* that corresponded to the first fermentor of the multistage CSTR system. Using a glucose concentration of 260 gl⁻¹ in the medium, the highest ethanol concentration reached was in batch (116 gl⁻¹), followed by the multistage CSTR (106 gl⁻¹), and the single stage CSTR

continuous production system (60 gl⁻¹). The highest ethanol productivity at this sugar concentration was achieved in the multistage CSTR system where a productivity of 12.7 g⁻¹h⁻¹ was seen. The other fermentation systems in comparison did not exceed an ethanol productivity of 3 g⁻¹h⁻¹. By performing a

continuous ethanol fermentation in multiple stages (having a total equivalent working volume of the tested single stage), a 4-fold higher ethanol productivity was achieved as compared to either the single stage CSTR, or the batch fermentation.

4. E. Bellissimi and W.M. Ingledew. 2005. Metabolic acclimatization: preparing active dry yeast for fuel alcohol production. Process Biochem **40**:2205-2213.

"Propagation" or "conditioning" of active dry yeast (ADY) for the production of fuel ethanol is thought to reduce lag times in fermentation and reduce overall fermentation times. The objectives of this study were to determine the optimal time that ADY should be conditioned prior to fermentation and to determine how well yeast and bacterial contaminants present in ADY are propagated. A new term, metabolic acclimatization (MAcc), was introduced to describe events occurring during conditioning. It was observed that MAcc is not necessary prior

to alcoholic fermentation. Lag times were reduced but overall fermentation times were unaffected. In fact, fermentation rates were slower than control conditions where ADY was directly inoculated into fermentors. In addition, if MAcc systems were to be used, it is recommended that only batch MAcc systems be practiced. Continuous MAcc (continuous yeast propagation) leads to increased bacterial numbers and organic acid levels which become stressful to yeast.

5. D. A. Abbott and W.M. Ingledew. 2005 Growth rates of *Dekkera/Brettanomyces* yeasts hinder their ability to compete with *Saccharomyces cerevisiae* in batch corn mash fermentations. Appl Microbiol Biotechnol **66**:641-647.

Growth rates determined by linear regression analysis revealed that *Saccharomyces cerevisiae* consistently grew more rapidly than *Brettanomyces* yeasts under a wide array of batch fermentative conditions, including acetic acid stress, in normal gravity (~20°P) mashes made from ground corn. *Brettanomyces* yeasts only grew more rapidly than *Saccharomyces cerevisiae* when acetic acid concentrations were elevated to industrially irrelevant levels (> 0.45% w/v). Furthermore, the 3 *Brettanomyces* isolates used in this study failed to produce significant quantities of acetic acid under pure culture fermentative conditions. In fact, the small amounts of acetic acid which accumulated in pure culture fermentations of whole corn mash were below the concentration required to inhibit the growth and metabolism of *Saccharomyces cerevisiae*. Acetic acid concentrations in pure culture *Brettanomyces* fermentations exceeded 0.05% w/v only in media containing low levels of glucose (<4% w/v) or when aeration rates were elevated to at least 0.03 vol air/vol mash/min. Consequently, it was concluded that *Brettanomyces* yeasts would not be capable of competing with *Saccharomyces cerevisiae* in industrial batch fermentations of whole corn mash based solely on growth rates, nor would they be capable of producing inhibitory concentrations of acetic acid in such fermentations.

VI. Department of Genetics, University of Debrecen, POBox 56, 4010 Debrecen, Hungary. Communicated by Matthias Sipiczki - <u>lipovy@tigris.unideb.hu</u>

List of recent papers.

- 1. Sipiczki, M., Romano, P., Capece, A., Paraggio, M. 2004. Genetic segregation of natural *Saccharomyces cerevisiae* strains derived from spontaneous fermentation of Aglianico wine. J. Appl. Microbiol. **96**:1169-1175.
- 2. Sipiczki, M. 2004. Species identification and comparative molecular and physiological analysis of *Candida zemplinina* and *Candida stellata*. J. Basic. Microbiol. 44:471-479.
- 3. Sipiczki, M. 2004. Fission Yeast Phylogenesis and Evolution. In "Molecular Biology of *Schizosaccharomyces pombe*" (Ed. R. Egel) Springer Verlag, Heidelberg, pp. 431-443.
- 4. Czako-Ver, K., Koosz, Z., Antal, J., Racz, T., Sipiczki, M., Pesti, M. 2004. Characterization of chromate-sensitive and -tolerant mutants of *Schizosaccharomyces pombe*. Folia Microbiol. 49:31-36.
- 5. Benko, Z., Fenyvesvolgyi, C., Pesti, M., Sipiczki, M. 2004. The transcription factor Pap1/Caf3 plays a central role in the determination of caffeine resistance in *S. pombe*. Mol. Genet. Genomics 271:161-170.
- Bourbon, H-M., Aguilera, A., Ansari, A., Asturias, F.J., Berk, A.J., Bjorklund, S., Blackwell, T.K., Borggrefe, T., Carey, M., Carlson, M., Conaway, J.W., Conaway, R.C., Emmons, S.W., Fondell, J.D., Freedman, S.W., Fukasawa, T., Gustaffson, C.M., Han, M., He, X., Herman, P.K., Hinnebush, A.G., Holmberg, S., Holstege, F.C., Jaehning, J.A., Kim, Y-J., Kuras, L., Leutz, A., Lis, J.T., Meisterernest, M., Naar, A.M., Nasmyth, K., Parvin, J.D., Ptashne, M., Reinberg, D., Ronne, H., Sadowski, I., Sakurai, H., Sipiczki, M., Sternberg, P.W., Stillman, D.J., Strich, R., Struhl, K., Svejstrup, J.Q., Tuck, S., Winston, F., Roeder, R.G., Kornberg, R.D. 2004. A unified nomenclature for protein subunits of Mediator complexes linking transcriptional regulators to RNA polymerase II. Molecular Cell 14:553-557.

- 7. Sugita, T., Takeo, K., Ohkusu, M., Virtudazo, E., Takashima, M., Asako, E., Ohshima, F., Harada, C., Nishikawa, A., Majoros, L., Sipiczki, M. 2004. Fluconazole-resistant pathogens *Candida inconspicus* and *C. norvegicus*: DNA sequence diversity of the rRNA intergenic spacer region, antifungal drug susceptibility and extracellular enzyme production. Microbiol. Immunol. 48:761-766.
- 8. Drivinya, A., Szilagyi, Z., Sipiczki, M., Takeo, K., Shimizu, K. 2004. Structural and functional analysis of genes encoding fork head proteins in *Cryptococcus neoformans*. Biologia. 56:711-718.
- 9. Szilagyi, Z., Batta, G., Enczi, K., Sipiczki, M. 2005. Characterisation of two novel fork-head gene homologues of *Schizosaccharomyces pombe*: Their involvement in cell cycle and sexual differentiation. Gene 348:101-109.
- 10. Alonzo-Nunez, M.L., An, H., Mehta, S., Petit, C., Sipiczki, M., del Rey, F., Gould, K.L., Vazquez de Aldana, C.R. 2005. Ace2p controls the expression of genes required for cell separation in *Schizosaccharomyces pombe*. Mol. Biol. Cell 16:2003-2017.
- 11. Antunovics, Z., Irinyi, L., Sipiczki, M. 2005. Combined application of methods to taxonomic identification of *Saccharomyces* strains in fermenting botrytized grape must. J. Appl. Microbiol. 98:971-979.
- 12. Lee, K.M., Miklos, I., Du, H., Watt, S., Szilagyi, Z., Saiz, J.E., Madabhushi, R., Sipiczki, M., Bahler, J., Fischer, R.P. 2005. Specific defects in cell cycle-regulated gene expression without general transcription shutdown in fission yeast Mcs6 complex (TFIIH-associated CDK-activating kinase) mutants. Mol. Biol. Cell, Apr 13; [Epub ahead of print].
- 13. Antunovics, Z., Nguyen, H-V., Gaillardin, C., Sipiczki, M. 2005. Gradual genome stabilisation by progressive reduction of the S. uvarum genome in an interspecific hybrid with S. cerevisiae. FEMS Yeast Res. (in press).
- 14. Willer, T., Brandl, M., Sipiczki, M., Strahl, S. 2005. Protein O-mannosylation is crucial for cell wall integrity, septation and viability in fission yeast. Mol. Microbiol. (in press).

VII. Department of Biology, Carleton University, Ottawa, Ontario, Canada K1S 5B6. Communicated by B.F. Johnson - <u>hb4johnson@primus.ca</u>

Recent publications

- 1. BF Johnson, M Bassan, & GB Calleja 2004 Anomalous death/viability patterns of two yeasts cultured in Yeast Nitrogen Base. Yeast Newsletter **53**:14.
- 2. BF Johnson, BY Yoo, GB Calleja & CP Kozela (In press) Second thoughts on septation by the fission yeast, *Schizosaccharomyces pombe*: Pull *vs*. push mechanisms. Antonie van Leeuwenhoek.

The correlation of contraction of an actomyosin band with the closing of the septum of the fission yeast cannot suggest cause-and-effect because contraction would be apparent whether the membrane enveloping the centripetally closing septum were pulled or were pushed. Thus the common observation of contraction is not critical. Diagrams of published electron micrographs of dividing wild-type fission yeasts illustrate variable (tilted) septal images that are counterintuitive to a pull model. Circumference calculations based on those images suggest that some variable forms might be only 6% closed even though their 2-dimensional profiles would be 50% closed, if they were not tilted. Development of multiseptate forms of cdc4-8 and *cdc4*-377 temperature sensitive mutants incubated at their restrictive temperature was followed. These multiseptate forms are shown to have functional (functional in terms of generating divided uninucleate cytoplasts) but grotesque septa which are

formed in the absence of actomyosin bands. By contrast, the myosin of the plant phragmoplast is not properly oriented for contractility, and *Dictyostelium* (attached cells) and *Saccharomyces* (mutants) have been shown to divide in the absence of myosin II, just as *Schizosaccharomyces* does (above). Hence contractility, the essence of a pull model for septum closure would seem to be non-essential. Other, non-contractile mechanisms of myosin are emphasized, and a push model becomes a rational default hypothesis. The essence of push models is that their synthesis/assembly mechanisms could be driving force sufficient for septum closure.

Appendix: Dimensional modelling of the flat and variable septa (BF Johnson, CP Kozela, P Novak, O Clarkin, N Wolters & S Cock).

VIII. Russian Collection of Microorganisms (VKM), Institute for Biochemistry and Physiology of Microorganisms, Russian Academy of Sciences, Pushchino 142292, Russia. Communicated by W.I. Golubev - wig@ibpm.pushchino.ru

Recent publications

1. Pfeiffer I, Golubev WI, Farkas Z, Kucsera J, Golubev N 2004 Mycocin production in *Cryptococcus aquaticus*. Antonie van Leeuwenhoek **86**:369-375.

Double-stranded RNA viruses of about 35 nm in diameter were isolated from a mycocin-secreting strain of *Cryptococcus aquaticus*. A derivative of this strain, lacking small dsRNA, was non-mycocinogenic and sensitive to its own toxin. The killing pattern of this mycocin was restricted to some species of the Cystofilobasidiales clade. Despite the differences in genome size of dsRNA viruses in mycocinogenic strains of *Cr. aquaticus, Cystofilobasidium* sp. CBS 6569, *Cyst. bisporidii, Cyst. infirmominiatum, Trichosporon pullulans* and *Xanthophyllomyces dendrorhous* and kiling patterns of their mycocins, the viral genomes showed homology in hybridisation experiments.

2. Golubev WI, Golubeva EW 2004 (In press) Yeast fungi in steppe and forest phytocenoses of the Prioksko-terrasny biosphere reserve. Mycologia i Phytopathologia **38**(6).

Using various media a survey was made in the biosphere Prioksko-terrasny reserve (Moscow region) of the yeasts living on herbaceous plants, in litter and soil during four seasons (1997-2000). Yeast communities from steppe and forest phytocenoses were compared. The yeasts isolated comprised more than 40 species belonging to the genera *Bullera*, *Candida*, *Cryptococcus*, *Cystofilobasidium*, *Debaryomyces*, *Dioszegia*, *Pichia*, *Pseudozyma*, *Rhodosporidium*, *Rhodotorula*, *Trichosporon* and *Udeniomyces*. The species *B. unica*, *B. huianensis*, *C. railenensis*, *C. santjacobensis*, *Cr. nemorosus*, *Cr. perniciosus, Ps. fusiformata, Rh. colostri, Tr. faecale, Tr. moniliiforme* and *Tr. porosum* were recovered for the first time in Russia. The most common yeasts were *B. unica, Cr. laurentii, Cr. magnus* from steppe plants and the species *C. railenensis, C. santjacobensis, C. wickerhamii, Cr. aerius, Cr. laurentii, Rh. auricularia* from forest litter. Killer yeasts were found at high frequency in the habitats of high density of yeast populations, namely, from phylloplane in steppe locality and from litter in deciduous forests.

3. Golubev WI, Sampaio JP, Alves L, Golubev NW 2004 *Cryptococcus festucosus* sp. nov. a new hymenomycetous yeast in the *Holtermannia* clade. Can J Microbiol **50**:1001-1006.

Five yeast strains belonging to the genus *Cryptococcus* Vuillemin were isolated from steppe plants and turf collected in the Prioksko-terrasny biosphere reserve (Moscow region, Russia). Sequence analyses of the D1/D2 domains of the 26S rDNA and of the internal transcribed spacer region revealed that these strains and the strain CBS 8016 have almost identical sequences and belong to the *Holtermannia* clade of the Tremellomycetidae (Basidiomycota, Hymenomycetes). A novel species named *Cryptococcus festucosus* (type strain VKM Y-2930) is proposed to accommodate these strains. Physiological characteristics and mycocin sensitivity patterns distinguishing *Cryptococcus festucosus* from the other species of this clade are presented.

4. Golubev WI, Golubeva EW 2005 *Quadrisporomyces adherentes* of the genus *Schizosaccharomyces*? Mykologia i Phytopathologia **39**:16-17.

Three strains described as *Quadrisporomyces adherentes* Sekunova were examined for their cultural, morphological and physiological characteristics, and this name

is placed in synonymy with *Schizosaccharomyces pombe* Lindner.

IX. Laboratorio de Microbiologia Aplicada y Biotecnologia, Universidad Nacional del Comahue, CRUB, Quintral 1250 (8400), Bariloche, RN, Argentina. Communicated by D. Libkind libkind@crub.uncoma.edu.ar

Publications in the last two years.

- 1. Libkind D, Brizzio S, Ruffini A, Gadanho M, van Broock MR, Sampaio JP 2003 Molecular characterization of carotenogenic yeasts from aquatic environments in Patagonia, Argentina. Antonie van Leeuwenhoek 84:313-322.
- 2. Libkind D, Pérez P, Sommaruga R, Diéguez MC, Ferraro M, Brizzio S, Zagarese H, van Broock MR 2004 Constitutive and UV-inducible synthesis of photoprotective compounds (carotenoids and mycosporines) by freshwater yeasts. Photochem Photobiol Sci 3:281-286.
- 3. Libkind D, Brizzio S, van Broock MR 2004 *Rhodotorula mucilaginosa*, a carotenoid producing yeast strain from a Patagonian high altitude lake. Fol Microbiol 49(1):19-25.
- 4. Sommaruga R, Libkind D, van Broock M, Whitehead K 2004 Mycosporine-glutaminol-glucoside, a UV-absorbing compound of two *Rhodotorula* yeast species. Yeast 12:1077-1081.

5. Libkind D, Gadanho M, van Broock, MR, Sampaio JP 2005 *Sporidiobolus longiusculus* sp. nov. and *Sporobolomyces patagonicus* sp. nov., novel yeasts of the Sporidiobolales isolated from aquatic environments in Patagonia, Argentina. Int J Syst Evol Microbiol 55:503-509.

Publications in press or submitted

6. Libkind D, Sommaruga R, Zagarese H, van Broock, MR (In press) Mycosporines in carotenogenic yeasts. Syst Appl Microbiol

We assessed the ability to produce mycosporines (MYCs) in 157 pigmented yeast strains (8 genera, 25 species) isolated from natural environments of Patagonia (Argentina). The strains belong to four taxonomic groups: the Sporidiobolales and *Erythrobasidium* clade of the class Urediniomycetes, and Cystofilobasidiales and Tremellales of the class Hymenomycetes. Induction of mycosporines did not occur in all yeast strains tested and appeared to be an exclusive trait of members of the

Erythrobasidium clade and Tremellales. This is the first report on the production of MYCs by pigmented species from the latter group, as well as the first extensive screening of mycosporinogenic yeasts. The consistent occurrence of MYCs in some specific phylogenetic groups suggests this trait bears evolutionary significance and that the presence/absence of MYC may have potential applications in yeast systematics.

X. Centre for Infectious Diseases and Microbiology, The University of Sydney, Level 3, Room 3114A, PO Box 533, Wentworthville, NSW 2145, Australia. Communicated by W. Meyer w.meyer@usyd.edu.au

Recent publication

1. SE Kidd, F Hagen, RL Tscharke, M Huynh, KH Bartlett, M Fyfe, L MacDougall, T Boekhout, KJ Kwon-Chung, and W Meyer 2004 A rare genotype of *Cryptococcus gattii* caused the cryptococcosis outbreak on Vancouver Island (British Columbia, Canada). Proc Nat Acad Sci 2004 101(49):17258-17263.

Cryptococcus gattii causes life-threatening infection of the pulmonary and central nervous systems in hosts with normal immunity, and has traditionally been considered to be geographically restricted to tropical and sub-tropical climate. The recent outbreak of *C. gattii* in the temperate climate of Vancouver Island, British Columbia (B.C.), Canada, led to a collaborative investigation. The objectives of the current study were to ascertain the environmental source of the outbreak and environmental cryptococcal isolates, and to determine the extent of genetic diversity among the isolates. PCR-fingerprinting and Amplified Fragment Length Polymorphism (AFLP) were used to examine the genotypes, and mating assays were performed to determine the mating type of the isolates. All outbreak and environmental isolates belonged to *C. gattii*. Concordant results were obtained using PCR-fingerprinting and AFLP analysis. The vast majority of clinical and veterinary infections were caused by isolates of the molecular type VGII/AFLP genotype 6, but two were caused by molecular type VGI/AFLP genotype 4. All environmental isolates belonged to molecular type VGI/AFLP genotype 6. Two/three subtypes were observed within VGII/AFLP6 among outbreak and environmental isolates. All mating competent isolates were of the alpha mating type. The emergence of this usually tropical pathogen on Vancouver Island highlights the changing distribution of this genotype, and emphasises the importance of an ongoing collaborative effort to monitor the global epidemiology of this yeast.

XI. Research Institute for Viticulture and Biology, Matúškova 25, 831 01 Bratialava, Slovakia. Communicated by E. Minárik.

The following are summaries of articles which had been recently published

1. Minárik E 2005 Active dry wine yeasts versus spontaneous grape must fermentation. Vinařský obzor 98:29 (in Slovak).

Reflections on the uniformity of wines fermented by active dry wine yeasts (ADWY) sporadically originate from conservative wine producers. Hundreds of scientific and professional publication in recent years point out the unique specialized character of selected ADWY cultures used in wineries worldwide with optimal wine quality. Examples are given in dry and semi-sweet white wines as well as in dry red wines exclusively fermenting with ADWY cultures. Best results are also attained in refermentations of stuck or sluggish fermentations and sparkling wine production.

Minárik E 2005 Importance of yeast ghost activators in grape must fermentation. Vinařský obzor 98:77 (in Slovak).

Yeast ghosts represent an outstanding aid attaining favourable results in grape must and sparkling wine fermentation even under unfavourable fermentation conditions, e.g., presence of laccase of botrytized grape berries, low fermentation temperature, etc. Yeast ghosts show positive influence on extending yeast viability. Recommended doses for grape must are 250-300 mg/L. It is underlined that microcrystalline cellulose displays similar activation properties.

3. Minárik E 2004 Importance of the yeast and bacterial flora on wine production. Vinič a víno 4: suppl. 3-4 (in Slovak).

The importance of inhibiting the indigenous microflora in grape must fermentation by pure yeast strains and selected strains of lactic acid bacteria (*Oenococcus oeni*) as soon as possible is underlined. in order to guarantee the metabiotic malolactic fermentation following the alcoholic fermentation. It is advantageous to inoculate *O. oeni* immediately after alcoholic fermentation is finished while the young wine is still on lees. Optimal conditions for both alcoholic and malolactic fermentations are recommended.

XII. Institut für Angewandte Mikrobiologie, Universität für Bodenkultur, Nuβdorfer Läende 11, A-1190 Vienna, Austria. Communicated by H. Prillinger - <u>hansjoerg.prillinger@boku.ac.at</u>

Recent publications

1. K Lopandic, O Molnár, M Suzuki, W Pinsker & H Prillinger (In press) Estimation of phylogenetic relationships within the Ascomycota on the basis of 18S rDNA sequences and chemotaxonomy. Mycological Progress.

Small subunit rRNA gene sequences (18S rDNA), cell wall carbohydrate composition and ubiquinone components were analysed within a larger number of ascomycetous yeasts and dimorphic fungi to validate their congruence in predicting phylogenetic relationships. The glucose-mannose pattern distinguishes the Hemiascomycetes from the Euascomycetes and the Protomycetes which are characterised with the glucose-mannose-galactose-rhamnose-(fucose) profile. The glucose-mannose-galactose pattern was found in the cell walls of all the three classes. Different coenzyme Q component (CoQ5 to CoQ10) were found within the representatives of the Hemiascomycetes. Whereas CoQ9, CoQ10 and CoQ10H2 predominate within the Euascomycetes, CoQ9 and CoQ10

characterise the Protomycetes. Chemotaxonomic studies coupled with additional molecular and co-evolution studies support the idea that the Hemiascomycetes occupy a basal position in the phylogeny of Ascomycota. These results are not in line with the phylogenetic studies based on the sequences of 18S rRNA encoding gene. The maximum parsimony analysis indicated that Hemiascomycetes and Protomycetes might represent sister groups, opposing to the earlier reported results, where the Archiascomycetes (Protomycetes) or the Hemiascomycetes had been considered to be the most primitive ascomycetous fungi. Instead of the class Archiascomycetes, the term Protomycetes was introduced reflecting much better the properties of the whole class.

2. M Wuczkowski & H Prillinger 2004 Molecular identification of yeasts from soils of the alluvial forest national park along the river Danube downstream of Vienna, Austria ("Nationalpark Donauauen"). Microbiol Res 159:263-275.

We analysed the diversity of yeasts from different soils in a river-floodplain landscape at the river Danube downstream of Vienna, Austria ("Nationalpark Donauauen"). 136 strains were isolated, identification of species was done with molecular methods. Partial sequencing of the 26S rRNA gene resulted in 36 different sequences, they could be assigned to 16 genera, apart from two sequence types (from three isolates) which were not clearly assigned to any genus. 18 species were identified and confirmed by means of PCR fingerprinting. The most frequently isolated genus was Cryptococcus (61 isolates and 12 sequence types). Basidiomycetes dominated with about 60 % above the members of the Ascomycetes. About half the yeasts was isolated from the litter, the quantity decreased with soil depth.

3. O Molnar & H Prillinger (In press) Analysis of yeast isolates related to *Metschnikowia pulcherrima* using the partial sequences of the large subunit rDNA and the actin gene; description of *Metschnikowia andauensis* sp. nov. System Appl Microbiol

Thirty-two yeast isolates were cultured from guts or excrements of 3 different pests of corn or from the stem of healthy corn. The strains were analyzed using MSP-PCR (micro/minisatellite-primed polymerase chain reaction), sequences of the D1/D2 region of the large subunit rDNA and a 979 bp long part of the actin gene (act-1). They seem to belong to three groups that are all sister groups of *Metschnikowia* pulcherrima, *M. fructicola* and *M. chrysoperlae*. A new species, *Metschnikowia andauensis* (HA 1657T) is described. In contrary to *M. pulcherrima* and *M. fructicola*, *M. andauensis* is welseparated in the act-1 phylogenetic tree too.

4. M Wuczkowski, K Sterflinger, GF Kraus, B Klug & H Prillinger 2003 Diversity of microfungi and yeasts in soils of the alluvial zone national park along the river Danube downstream of Vienna, Austria ("Nationalpark Donauauen"). Die Bodenkultur, Austrian J Agric Res 54:109-117.

The diversity of microfungi and yeasts in different soils in a river-floodplain landscape at the river Danube downstream of Vienna, Austria was analysed. Soil samples were taken from six different sites: under *Salix* sp.- and *Populus* sp. trees in a seasonally flooded and a not flooded forest and from agricultural fields with ecological and conventional farming. Material was taken from the litter and from three soil cores: 0-5, 10-15 and 30-35 cm. Fungi were identified morphologically to genus level, yeast strains by sequencing the D1-D2 region of the 26S-rDNA. The seasonally flooded and the not flooded forest showed no difference, but the diversity of microfungi was higher under *Populus* trees.

XIII. Sección Enología, Cátedra de Ciencia y Tecnología de Alimentos, Facultad de Química, Universidad de la República. Av. Gral. Flores 2124, 11.800 Montevideo, Uruguay. Comunicated by F.M. Carrau - <u>fcarrau@fg.edu.uy</u> - <u>http://enologia.fg.edu.uy</u>

The following are our most recent publications related to wine yeasts.

1. Gonzalez Techera A, S Jubany, FM Carrau, C Gaggero 2001 Differentiation of industrial wine yeast strains using microsatellite markers. Lett Appl Microbiol UK **33**:71-75.

Aims: To differentiate nine industrial wine strains of *Saccharomyces cerevisiae* using microsatellite (simple sequence repeats, SSR) markers. Methods and Results: Six of the strains were indigenous yeasts currently used as highdensity starter monocultures by the Uruguayan wine industry. Unequivocal differentiation of these six native strains and three commercial *S. cerevisiae* wine strains was achieved by PCR amplification and polymorphism analysis of loci containing microsatellite markers.

Conclusions: We recommend the use of this reproducible and simple molecular method to routinely discriminate wine yeast strains. Significance and Impact of the Study: Microsatellites are superior to other methods for typing yeasts because the results can be exchanged as quantitative data. Knowledge of the frequencies of the alleles for different SSR markers will eventually lead to an accurate typing method to identify industrial wine yeast strains.

 Carrau FM, Medina K, Boido E, Farina L, Gaggero C, Dellacassa E, Versini G, Henschke PA 2005 De novo synthesis of monoterpenes by *Saccharomyces cerevisiae* wine yeasts. FEMS Microbiol Lett 243:107–115

This paper reports the production of monoterpenes, which elicit a floral aroma in wine, by strains of the yeast *Saccharomyces cerevisiae*. Terpenes, which are typical components of the essential oils of flowers and fruits, are also present as free and glycosylated conjugates amongst the secondary metabolites of certain wine grape varieties of *Vitis vinifera*. Hence, when these compounds are present in wine they are considered to originate from grape and not fermentation. However, the biosynthesis of monoterpenes by *S. cerevisiae* in the absence of grape derived precursors is shown here to be of de novo origin in wine yeast strains. Higher concentration of assimilable nitrogen increased accumulation of linalool and citronellol. Microaerobic compared with anaerobic conditions favored terpene accumulation in the ferment. The amount of linalool produced by some strains of *S. cerevisiae* could be of sensory importance in wine production. These unexpected results are discussed in relation to the known sterol biosynthetic pathway and to an alternative pathway for terpene biosynthesis not previously described in yeast.

3. Medina K, Boido E, Dellacassa E, Carrau FM 2005 Yeast interactions with anthocyanins during red wine fermentation. Am J EnolVitc **56**:104-109.

Yeast capacity to modify anthocyanin content during red wine fermentation was studied. Our main objective was the selection of yeast strains according to their capacity for color removal during vinification. A model red grape juice medium was developed and evaluated to better understand yeastanthocyanin interactions. An anthocyanin extract of *Vitis vinifera* cv. Tannat was prepared from grape skins and diluted in a white must juice in order to obtain a red grape juice without solids. Anthocyanin removal was confirmed to be higher for compounds with higher polarity. Acylated anthocyanins (acetyl and *p*-coumaryl compounds) were differentially removed, and the

The following paper was recently submitted.

4. Rabosto X, Carrau M, Paz A, Boido E, Dellacassa E, Carrau FM Grapes and vineyard soils as sources of microorganisms for biological control of *Botrytis cinerea*.

Biological control agents are becoming increasingly interesting as alternatives to chemical fungicides, which may be hazardous to the environment and can promote resistance in pathogens. Wine grape cuticles of several varieties and soil of different Uruguayan vineyards were checked for the presence of microorganisms, which could be antagonistic to the phytopathogenic fungus *Botrytis cinerea*. From 223 isolates of yeasts and bacteria, only 8 non-Saccharomyces yeast strains and 4 bacteria showed more than 50% effectiveness index against Botrytis cinerea in vitro. These strains represent less than 5% of the total number of microorganisms naturally present in grapes. Bacillus sp. isolate UYBC38 and a yeast Hanseniaspora uvarum isolate UYNS13, showed high antagonistic capability against the pathogen and both were very effective (100% and 90% respectively) in controlling rot development on grape clusters. These two strains were selected for further studies in lab and vineyard applications. Before field trials both strains were tested

percentage of removal for each compound was determined. Results obtained showed no correlation between color intensity and total anthocyanin content after fermentation, which was explained by the presence of anthocyanin derivates formed during the process. HPLC-MS analysis allowed identification of anthocyanin derivates, while the quantification of several derivatives was performed by HPLC-DAD. In addition, the sum of these derivates showed a direct correlation with the color intensity obtained with each strain, thus explaining the color variability observed.

against the wine yeast M522 and other natural non-Saccharomyces strains so as to prevent fermentation problems. The combined mechanisms of antagonism that may contribute to biocontrol were studied against B. cinerea. Yeast strain UYNS13 showed only competition for nutrients. Although the Bacillus sp strain also consume significantly more nitrogen from a grape juice than B. cinerea, the relevant mechanism of antagonism of this bacteria was the production of antifungal substances that completely inhibited development of B. cinerea conidia in vitro, evidencing disruption of spore cell wall and leakage of protoplasm. Preliminary commercial application of UYBC38 in the vineyard shows that this strain could survive in leaves and fruits between 45 and 70 days after the sprays. The strategy of increasing the natural antagonistic population present in vineyards to prevent fungal diseases is discussed as a "low input viticulture" practice.

XIV. Botanisches Institut der Universitäät Dusseldorf, Universitatsstraßße 1, D-40225 Dusseldorf, Germany. Communicated by Georg Michaelis - michaelg@uni-duesseldorf.de

Recent publications.

- 1. Esser K, Tursun B, Ingenhoven M, Michaelis G, Pratje E 2002 A novel two-step mechanism for removal of a mitochondrial signal sequence involves the mAAA complex and the putative rhomboid protease Pcp1. J Mol Biol 323:835-843.
- 2. K Esser, PS Jan, E Pratje, G Michaelis 2004 The mitochondrial IMP peptidase of yeast: functional analysis of domains and identification of Gut2 as a new natural substrate. Mol. Gen Genet 271:616-626.

The mitochondrial inner membrane peptidase IMP of *Saccharomyces cerevisiae* is required for proteolytic processing of certain mitochondrially and nucleus-encoded proteins during their export from the matrix into the inner membrane or the intermembrane space. The membrane associated signal peptidase complex is composed of the two catalytic subunits Imp1 and Imp2 and the Som1 protein. The IMP subunits may exert functions in membrane association, interaction and stabilisation of subunits, substrate specificity, and proteolysis. We have analysed inner membrane peptidase mutants and substrates to gain more insight into the function of various domains and substrate recognition. The data suggest that certain conserved glycine residues in the second and third conserved regions in Imp1 and Imp2 are important for stabilisation of the Imp complex or for proteolytic activity of the subunits, respectively. The non-

conserved C-terminal parts of the Imp subunits are important for their proteolytic activities. The C-terminus of Imp2, comprising a predicted second transmembrane segment, is dispensable for stabilisation of Imp2 or Imp1, and cannot substitute the function of the Imp1 C- terminus. Changing the Imp2 cleavage site of cytochrome c_1 (A⁺M) to (N⁺D) reveals a sequence specificity of the Imp2 peptidase. In addition we identified mitochondrial Gut2, the mitochondrial FAD- dependent glycerol-3-phosphate dehydrogenase, as a new Imp1 substrate, whose uncleaved precursor may contribute to the *pet* phenotype of certain imp mutants. Gut2 is associated to the inner membrane and is essential for growth on glycerol containing media. Suggested functions of the analysed residues and domains of the IMP subunits, characteristics of the cleaved sites of substrates and implications for the phenotypes of imp mutants are discussed.

- 3. K Esser, G Michaelis, E Pratje 2005 Extranuclear inheritance: mitochondrial genetics and biogenesis. Progress in Botany 66:91-111.
- 4. G Michaelis, K Esser, B Tursun, JP Stohn, S Hanson, E Pratje 2005 (in press) Mitochondrial signal peptidases of yeast: the rhomboid peptidase Pcp1 and its substrate cytochrome c peroxidase. Gene

The rhomboid peptidase Pcp1 of yeast is the first mitochondrial enzyme of this new class of serine peptidases. Pcp1 is an integral part of the inner membrane and was identified by its signal peptidase activity responsible for processing of the intermediate of cytochrome c peroxidase (iCcp1) to the mature enzyme (Esser et al. 2002). Here we describe studies on the

expression of the *PCP1* gene. Proteolytic processing of Pcp1 itself was found. The precursor and the intermediate of Ccp1 were localized to the inner membrane. The results confirm our previous report on a two step processing pathway of cytochrome c peroxidase and the identification of the signal peptidases involved.

XV. Collection de Levures d'Intérêt Biotechnologique (Clib), Laboratoire de Microbiologie et Génétique Moléculaire, INA-PG INRA, BP01, F-78850 Thiverval-grignon, France. Communicated by Nguyen H.V. - <u>nguyenhv@grignon.inra.fr</u>

The following have been recently published.

1. HV Nguyen and C Gaillardin 2005 Evolutionary relationships between the former species *Saccharomyces uvarum* and the hybrids *Saccharomyces bayanus* and *Saccharomyces pastorianus*; reinstatement of *Saccharomyces uvarum* (Beijerinck) as a distinct species. FEMS Yeast Res **5**:315-498.

Abstract. Analysis of the nucleotide sequence of the *GDH1* homologues from *Saccharomyces bayanus* strain CBS 380^T and *S. pastorianus* strains showed that they share an almost identical sequence, *SuGDH1**, which is a diverged form of the *SuGDH1* from the type strain of the former species *S. uvarum*, considered as synonym of *S. bayanus. SuGDH1** is close to but differs from *SuGDH1* by the accumulation of a high number of neutral substitutions designated as Multiple Neutral Mutations Accumulation (MNMA). Further analysis carried out with three other markers, *BAP2*, *HO* and *MET2* showed that they have also

Updated nomenclature. Differentiation of *S. uvarum* from *S. bayanus* and its reinstatement as species lead to the following changes in the naming of sequences issued from strains belonging to them:

1. Sequences obtained (a limited number) with strain CBS 380^T (CCRC 21960, CLIB 181, DBVPG 6171, DSM 70412,

diverged from their *S. uvarum* counterparts by MNMA. *S. bayanus* CBS 380^{T} is placed between *S. uvarum* and *S. pastorianus* sharing *MET2*, *CDC91* sequences with the former and *BAP2*, *GDH1*, *HO* sequences with the latter. *S. bayanus* CBS 380^{T} has been proposed to be a *S. uvarum/S. cerevisiae* hybrid and this proposal is confirmed by the presence in its genome a *S. cerevisiae SUC4* gene. Strain *S. bayanus* CBS 380^{T} , with a composite genome, is genetically isolated from strains of the former *S. uvarum* species, thus justifying the reinstatement of *S. uvarum* as a distinct species.

IFO 1127, IGC 4456, JCM 7258, NRRL Y-12624) labelled *S. bayanus* retain this name.

2. Sequences accessions from Yeast Comparative Genomics or Genolevures and obtained from strains MCYC623 and 623-6c should be reassigned to *S. uvarum* as these two strains belong to *S.uvarum*, similar to strain CBS 395^{T} (CCRC

21970; CLIB 251, DBVPG 6179, DSM 70547, IFO 0615, IGC 4567, NCYC 509, NRRL Y-12663, NRRL Y-17034)

(http://www.ncbi.nih.gov/mapview/map_search.cgi?taxid=226127) http://cbi.labri.u-bordeaux.fr/Genolevures/

3. The two strains MCYC623 and 623-6c used in three separate Genome projects are almost the same. MCYC623 was first described as *S. abuliensis* by Santa Maria (Microbiological Collection of Yeast Cultures, Department of Microbiology in the Agronomic School of Madrid, Spain), this strain was also deposited in the Centraalbureau voor Schimmelcultures as CBS (CLIB 533) is an *ura*3-1 mutant selected from a monosporic culture issued from strain MCYC623. Consequently the genome of *S. uvarum* strain CBS 7001 (MCYC623) is covered about 11X by the two sequencing projects of Cliften et al. (2003) Washington University School of Medecine, and Kellis et al. (2003). In 1991 the yeast strains of the MCYC (not to be confused with NCYC) were transferred to the Collection Española de Cultivo Tipo (CECT; <u>http://www.cect.org</u>)].

7001 (CLIB 283, DBVPG 6299, NRRL Y-11485). Strain 623-6c

2. Z Antunovics, HV Nguyen, C Gaillardin, M Sipiczki (In press) Gradual genome stabilisation by progressive reduction of the *S. uvarum* genome in an interspecific hybrid with *S. cerevisiae*. FEMS Yeast Res

Considerable amount of molecular and genetic data indicate that interspecific hybridisation may not be rare among natural strains of *Saccharomyces* sensu stricto. Although a postzygotic barrier operating during meiosis usually prevents the production of viable spores, stable hybrids can arise which can even evolve into distinct species. This study was aimed to analyse the genome of a fertile *Saccharomyces cerevisiae* x *S. uvarum* hybrid and monitor its changes over four filial generations of viable spores. The molecular genetic analysis demonstrated that the two species did not contribute equally to the formation and stabilisation of the hybrid genome. *S. cerevisiae* provided the mitochondrial DNA and the more stable part of the nuclear genome. The *S. uvarum* part of the hybrid nuclear genome became progressively smaller by loosing complete chromosomes and genetic markers in the course of successive meiotic divisions. Certain *S. uvarum* chromosomes were eliminated and/or underwent rearrangements in interactions with *S. cerevisiae* chromosomes. Numerous *S. uvarum* chromosomes acquired *S. cerevisiae* telomere sequences. The gradual elimination of large parts of the *S. uvarum* genome was associated with a progressive increase of sporulation efficiency. We hypothesize that this sort of genomic alterations may contribute to speciation in *Saccharomyces* sensu stricto.

Others articles recently published by other laboratories in which the name S. uvarum is used.

Laboratoire de Microbiologie et de Technologie des Fermentations, UMR Sciences pour l'oenologie, INRA, 2 place Viala, F-34060 Montpellier Cedex 1, France.

Cheraiti N, Guezenec S, Salmon JM 2005 Redox interactions between *Saccharomyces cerevisiae* and *Saccharomyces uvarum* in mixed culture under enological conditions. Appl Environ Microbiol. **71**:255-260.

Wine yeast starters that contain a mixture of different industrial yeasts with various properties may soon be introduced to the market. The mechanisms underlying the interactions between the different strains in the starter during alcoholic fermentation have never been investigated. We identified and investigated some of these interactions in a mixed culture containing two yeast strains grown under enological conditions. The inoculum contained the same amount (each) of a strain of *Saccharomyces cerevisiae* and a natural hybrid strain of *S. cerevisiae* and *Saccharomyces uvarum*. We identified interactions that affected biomass, by-product formation, and fermentation kinetics, and compared the redox ratios of monocultures of each strain with that of the mixed culture. The redox status of the mixed culture differed from that of the two monocultures, showing that the interactions between the yeast strains involved the diffusion of metabolite(s) within the mixed culture. Since acetaldehyde is a potential effector of fermentation, we investigated the kinetics of acetaldehyde production by the different cultures. The *S. cerevisiae-S. uvarum* hybrid strain produced large amounts of acetaldehyde for which the *S. cerevisiae* strain acted as a receiving strain in the mixed culture. Since yeast response to acetaldehyde involves the same mechanisms that participate in the response to other forms of stress, the acetaldehyde exchange between the two strains could play an important role in inhibiting some yeast strains and allowing the growth of others. Such interactions could be of particular importance in understanding the ecology of the colonization of complex fermentation media by *S. cerevisiae*.

Laboratoire Vigne Biotechnologie et Environnement de l'Universite de Haute-Alsace, Colmar, France.

Demuyter C, Lollier M, Legras JL, Le Jeune C 2004 Predominance of *Saccharomyces uvarum* during spontaneous alcoholic fermentation, for three consecutive years, in an Alsatian winery. Appl Microbiol. **97**:1140-1148.

The purpose of this study was to determine the origin of the yeasts involved in the spontaneous alcoholic fermentation of an Alsatian wine. During three successive years, must was collected at different stages of the winemaking process and fermented in the laboratory or in the cellar. *Saccharomyces* yeasts were sampled at the beginning and at the end of the fermentations. *Saccharomyces cerevisiae* clones were genetically characterized by inter-delta PCR. Non-*S. cerevisiae* clones were identified as *Saccharomyces uvarum* by PCR-RFLP on *MET2* gene and characterized at the strain level by karyotyping. The composition of the *Saccharomyces* population in the vineyard, after crushing and in the vat was analyzed. This led to three main results. First, the vineyard *Saccharomyces* population was rather homogeneous. Second, new non-resident strains had appeared in the must during the winemaking process. Finally, the yeast population in the vat only consisted in *S. uvarum* strains. This 3-year study has enabled us to show the involvement of indigenous *S. uvarum* in the alcoholic fermentation.: This study gives a first insight into the polymorphism of *S. uvarum* strains involved in a spontaneous alcoholic fermentation.

Corrigendum.

B Dujon ... JL Souciet 2004 Genome evolution in yeasts. Nature 430:35-44.

The complete genome of *Yarrowia lipolytica* was obtained from strain CLIB 122 and not CLIB 99 as cited in Table 1 of the article. Consequently, sequences available in the

database (*i.e.*, NC_006072 *Yarrowia lipolytica* CLIB99 chromosome F, complete sequence in NCBI) should be relabelled as *Y. lipolytica* CLIB122.

XVI. CREM – Centro de Recursos Microbiológicos, Secção Autónoma de Biotecnologia, Faculdade de Ciências e Tecnologia, Universidade Nova de Lisboa, 2829-516 Caparica, Portugal. Communicated by J. P. Sampaio - jss@fct.unl.pt

The following papers have been recently published (abstracts were included in the last issue).

- 1. Gadanho M and Sampaio JP 2004 Application of temperature gradient gel electrophoresis to the study of yeast diversity in the estuary of the Tagus river, Portugal. FEMS Yeast Res 5:253-261.
- 2. Libkind D, Gadanho M, van Broock Mand Sampaio JP 2005 *Sporidiobolus longiusculus* sp. nov. and *Sporobolomyces patagonicus* sp. nov., two novel yeasts of the Sporidiobolales isolated from aquatic environments in Patagonia, Argentina. Int J Syst Evol Microbiol **55**:503-509.
- 3. Golubev WI, Sampaio JP, Alves L and Golubev NW 2005 *Cryptococcus festucosus* sp. nov. a new hymenomycetous yeast in the *Holtermannia* clade. Can J Microbiol **50**:1001-1006.

The following paper has been accepted for publication.

4. Almeida, JMGCF 2005 Yeast community survey in Tagus estuary. FEMS Microbiol Ecol

The yeast community in the waters of the Tagus estuary, Portugal, was followed for over a year in order to assess its dynamics. Yeast occurrence and incidence were measured and this information was related to relevant environmental data. Yeast occurrence did not seem to depend upon tides, but river discharge had a dramatic impact both on the density and diversity of the community. The occurrence of some yeasts was partially correlated with faecal pollution indicators. Yeast isolates were characterized by MSP-PCR fingerprinting and rDNA sequencing. The principal species found were *Candida* catenulata, *Candida intermedia*, *Candida parapsilosis*, *Clavispora lusitaniae*, *Debaryomyces hansenii*, *Pichia guilliermondii*, *Rhodotorula mucilaginosa* and *Rhodosporidium diobovatum*. The incidence of these species was evaluated against the environmental context of the samples and current knowledge about the substrates from which they are usually isolated.

The following PhD thesis was recently defended.

5. M. Gadanho. Polyphasic taxonomy and molecular characterization of yeast diversity from aquatic environments.

The work presented in this thesis is divided in three parts. The first part concerns the taxonomic re-evaluation of a group of pigmented yeasts belonging to the Sporidiobolales and related to *Rhodotorula glutinis* (chapters 2, 3 and 4). A polyphasic approach including phenotypic and genetic methods was applied and the species boundaries of *Rhodosporidium kratochvilovae* and *Rh. glutinis* were redefined. Several strains identified in the past as *Rh. glutinis* were reclassified in other species, including in a new species, *R azoricum*. Therefore, a more stable and reliable classification system was obtained for this group of yeasts. Another result obtained during this part of the work was the optimization of a method based on the PCR amplification of satellite DNA (MSP-PCR – micro/mini-satellite primed PCR). This technique proved to be a suitable method for a quick and reliable typing of large numbers of strains.

The second part of this work consisted in the study of yeast diversity in three different aquatic environments: (i) marine costal waters off the city of Faro in the south of Portugal; (ii) hydrothermal vents at the Mid-Atlantic Ridge near the Azores archipelago; and (iii) acidic and metal-rich waters of the Iberian Pyrite Belt (IPB) (chapters 5, 6 and 7). The strategy employed in this part of the work was based on the previously optimized MSP-PCR technique. Approximately 750 yeasts were isolated from the three aquatic environments and identified down to the species level. Species identifications were based on the comparison of the obtained MSP-PCR fingerprints with reference profiles or on sequence analysis of the DID2 domains of the 26S rDNA. The results obtained allowed the characterization of the

yeast communities of the three environments. A considerable number of isolates could not be identified probably because they represent new species, most of them affiliated with the Basidiomycetes.

In the third part of this thesis, culture-independent techniques were tested and optimized for the assessment of yeast (and other fungi) diversity. Special attention was given to the DNA extraction protocol from aquatic samples and to the temperature gradient gel electrophoresis (TGGE) method. The yeast diversity of the estuary of the Tagus river was investigated in chapter 8. The extreme environments of the IPB were studied in chapter 9 and a comparison between the culture-dependent (chapter 7) and the culture-independent approaches was performed. Moreover, the molecular approach used in the IPB study included the characterization of the entire microeukaryotic community. The results obtained suggested that yeasts constitute a small fraction of the microeukaryotic community. This situation is likely to limit the utilization of culture-independent approaches for the characterization of yeasts in natural aquatic environments. Nevertheless, it was possible to detect the dominant yeast populations without cultivating them. Moreover, some of the IPB yeasts detected through TGGE and cloning were not found when the culture-dependent approach was employed. On the other hand, the majority of IPB yeasts detected by cultivation procedures was not detected by TGGE / cloning. These results indicate that yeast diversity studies should combine both culturedependent and culture-independent methods.

XVII. Department of Biology, University of Western Ontario, London, Ontario, Canada N6A 5B7. Communicated by M.A. Lachance - <u>lachance@uwo.ca</u>

The following papers have now appeared in print.

- 1. Pimenta RS, PDD Alves, A Corrêa Jr, MA Lachance, GS Prasad, R Rajaram, BRRP Sinha and CA Rosa. 2005 *Geotrichum silvicola*, a novel asexual arthroconidial yeast species related to genus *Galactomyces*. Int. J. Syst. Evol. Microbiol. **55**: 497- 501.
- 2. Anderson TM, MA Lachance and WT Starmer 2004 The relationship of phylogeny to community structure: the cactus-yeast community. Amer Naturalist **164**:709-721.
- 3. Lachance MA & JM Bowles 2004 *Metschnikowia similis* sp. nov. and *Metschnikowia colocasiae* sp. nov., two ascomycetous yeasts isolated from *Conotelus* spp. (Coleoptera: Nitidulidae) in Costa Rica. Studies in Mycology **50**:69-76.
- 4. Lachance MA, CP Ewing, JM Bowles & WT Starmer 2005 *Metschnikowia hamakuensis* sp. nov., *Metschnikowia kamakouana* sp. nov., and *Metschnikowia mauinuiana* sp. nov., three endemic yeasts from Hawaiian nitidulid beetles. Int J Syst Evol Microbiol **55**:1369-1377.

Three heterothallic, haplontic yeast species, *Metschnikowia hamakuensis, Metschnikowia kamakouana* and *Metschnikowia mauinuiana*, are described from isolates associated with endemic nitidulid beetles living on various endemic plants on three Hawaiian islands. As morphospecies, they are similar to *Metschnikowia hawaiiensis*, but based on mating compatibility and ascospore formation, they can be assigned clearly to distinct biological species. Analysis of ITS/5.8S and D1/D2 large subunit rDNA sequences shows that, with *M. hawaiiensis* and two other isolates, these species form a distinct subclade within the large-spored *Metschnikowia* species, indicating that they are Hawaiian endemics. Type cultures are: *M. hamakuensis*, UWOPS04-207.1^T = CBS 10056^T = NRRL Y-27834^T (type, h⁺) and UWOPS 04-204.1 = CBS10055 = NRRL Y-27833 (allotype, h⁻); *M. kamakouana*, UWOPS 04-112.5^T = CBS10058^T = NRRL Y-27836T (type, h⁺) and UWOPS 04-109.1 = CBS 10057 = NRRL Y-27835 (allotype, h⁻); and *M. mauinuiana*, UWOPS 04-190.1^T = CBS 10060^T = NRRL Y-27838^T (type, h⁺) and UWOPS 04-110.4 = CBS 10059 = NRRL Y-27837 (allotype, h⁻).

Papers in press.

5. Rosa CA, Lachance MA, Pimentel M, Antonini Y, and Martins R. In Press. *Candida riodocensis* and *Candida cellae*, two new yeast species from the *Starmerella* clade associated with solitary bees in the Atlantic Rain Forest of Brazil. FEMS Yeast Res

Two new ascomycetous yeast species belonging to the *Starmerella* clade were discovered in nests of two solitary bee species in the Atlantic rain forest of Brazil. *Candida riodocensis* was isolated from pollen-nectar provisions, larvae and fecal pellets of nests of *Megachile* sp., and *Candida cellae* was found in pollen-nectar provisions of *Centris tarsata*. Analysis of the

sequences of the D1/D2 large-subunit ribosomal DNA showed that *C. riodocensis* is phylogenetically related to *C. batistae*, and the closest relative of *C. cellae* is *C. etchellsii*. The type strains are *C. riodocensis* UFMG-MG02 (= CBS $10087^{T} = NRRL$ Y-27859^T) *C. cellae* UFMG-PC04 (= CBS $10086^{T} = NRRL$ Y-27860^T).

6. Ruivo CCC, Lachance MA, Rosa CA, Bacci M & Pagnocca FC. In Press. *Candida bromeliacearum* sp. nov. and *Candida ubatubensis* sp. nov. two yeasts species isolated from the water tank of *Canistropsis seidelii* (Bromeliaceae). Int J Syst Evol Microbiol.

Two new yeasts species, *Candida bromeliacearum* and *Candida ubatubensis*, were isolated from the bromeliad tank of *Canistropsis seidelii* (Bromeliaceae) in a sandy coastal plain (restinga) ecosystem site at an Atlantic rainforest of southeastern Brazil. These species were genetically isolated from all other currently accepted ascomycetous yeasts based on their sequence

divergence in the D1/D2 domain of the large subunit rDNA and in the small subunit rDNA. The species occupy basal positions in the Metschnikowiaceae. The type strains are *Candida bromeliacearum* UNESP $00-103^{T}$ (= CBS 10002^{T} = NRRL Y-27811^T) and *Candida ubatubensis* UNESP $01-247R^{T}$ (= CBS 10003^{T} = NRRL Y-27812^T).

7. Morais PB, MA Lachance and CA Rosa. In press. *Saturnispora hagleri* sp. nov., a yeast species isolated from *Drosophila* flies in Atlantic Rain Forest in Brazil. Int J Syst Bacteriol

Six strains of a new yeast species belonging to the genus *Saturnispora* were isolated from two species of the *Drosophila fasciola* subgroup (*repleta* group) in an Atlantic Rain Forest site in Rio de Janeiro State, Brazil. Four strains were isolated from crops and one from external parts of *Drosophila*

cardinae. One came from external parts of *D. fascioloides*. Analysis of the sequences of the D1/D2 large-subunit ribosomal DNA showed that the new species is closely related to *S. dispora* and was described as *Saturnispora hagleri*. The type culture is UFMG-55^T (= CBS10007^T = NRRL Y-27828^T).

Obituary Martin W. Miller 1925-2005



Martin Wesley Miller, Professor Emeritus of the Food Science and Technology department at the University of California, Davis, died after a short illness on April 23, 2005, in Sacramento, CA. He was 80. A graveside service was held at the Davis Cemetery on May 5, followed by a reception at Sudwerk in Davis.

Born January 8, 1925 in Belden, Nebraska, to Emma B. and Dr. Earl E. Miller, his early years were spent in Scotia and Culbertson, Nebraska. His mother died when he was seven years old, and his father, the town doctor, died in 1939. He and his younger brother lived in foster homes until his graduation from high school.

Miller tried to enlist in the armed services when Pearl Harbor was attacked in 1941, but was refused because he was underage. On the night he graduated from Culbertson High School in May 1942, he turned down a scholarship to the University of Nebraska and took a job overseeing a rail shipment of cattle to California, where he joined his half-sister Mae. There Miller worked various jobs until he found a Navy recruiter who looked the other way, and he enlisted in the U.S. Navy, where he served as a radioman first class stationed in Honolulu until his discharge in 1946.

Following his military service, Miller entered UC Berkeley as a freshman and earned an A.B. in bacteriology (1950) and M.S. in food science (1952). It was here he

married Patricia Meyer in 1948 when both were students. After receiving his M.S. the family moved to West Sacramento so he could continue working on his doctorate, working closely with Emil Mrak in the new Department of Food Science and Technology at UC Davis.

Miller received a Ph.D. in microbiology in 1958, and the family moved to Davis, where he resided until his death. He joined the faculty of the Department of Food Science and Technology as an Assistant Professor in 1959 and rose through the academic ranks. An internationally recognized expert in food science, Miller authored two books and over 150 scientific articles in the fields of food dehydration, food microbiology, fermentation, food spoilage by yeasts and molds, yeast taxonomy and ecology.

The California dried fruit and olive industries sought Miller's advice as a researcher and consultant, and he was involved in international program activities in food science in Brazil, Saudi Arabia, and Egypt. In 1964 he received a Fulbright Senior Research Award to Australia and later the National Science Foundation Japan Society Science Award. He was elected a Fellow of the American Academy of Microbiology (1967) and of the Institute of Food Technologists (1981).

In addition to his research, throughout his 30-year academic career Miller loved working with students and was a popular lecturer. He also served as master advisor for the food science undergraduate major for many years, mentoring both undergraduate and graduate students from many countries.

After Miller officially retired from the university in 1988 he was recalled for several years to teach upper division and graduate bacteriology and mycology courses, for which he was uniquely qualified. He remained in touch with many of his former students until his death.

Miller loved the outdoors and was an avid fisherman and camper. Early in their marriage, he and Patricia often backpacked in Yosemite and climbed Half Dome. He also enjoyed playing cribbage and became involved with genealogy, tirelessly researching his own family's history and Patricia's as well.

Colleagues, students, friends and family will remember Miller for many things, including his pipe, his whistle, his cheerfulness and his sense of humor.

Miller is survived by his wife of 56 years, Patricia Miller, their son Stephen Miller, daughter Susan Hoffman and her husband Jeff, and grandchildren Melissa Miller, Heather Hoffman, Stephanie Hoffman and Jason Hoffman. His brother Stephen and many nieces and nephews also survive him. He was preceded in death by his half sister Mae and his son Craig.

The family requests that remembrances be made to the Friends of the UC Davis Arboretum, One Shields Avenue, UC Davis, Davis, CA 95616, or to a charity of the donor's choice.

Thanks to Teri Wolcott (UC Davis) and to the Davis Enterprise for their assistance in making this text available.

Reflections on Our Friend and Yeast Comrade Marty Miller

Although the shadows cast by the larger trees of the forest normally hinder the growth of the other vegetation, Mary Miller was definitely an exception to the rule. In spite of the gigantic "shadow" of our dearly remembered "yeast guru" (and long-time Editor of the Yeast Newsletter) Herman Phaff, Marty was able to carve out a significant scientific niche both in yeast taxonomy and industrial microbiology. His activities for the better half of a century as an industrial mycologist not only brought prestige to the UCD Food Science Department, but these activities more often than not provided important funding for the UCD yeast collection which now bears Herman Phaff's name.

Marty was also a born teacher, and many of his former students (among them AEV) greatly appreciated his (and his wife Pat's) friendliness, patience and guidance during their studies. His approach to teaching was more like a game that he was generously allowing you to play, as he made students feel like yeast taxonomy was so much fun that he just couldn't keep it all to himself. How many professors will stand over a microscope and tell you "Look! That's sex!"

We (AEV & ASM) fondly remember Marty's visits to our lab in Perugia, and appreciate the fact that our students were able to see that great "names" can be extremely human. In addition, we will always be grateful for the good times spent, yes, discussing yeast, but also simply enjoying a good Italian meal accompanied by one of the most famous products of Saccharomyces cerevisiae - of which Mary definitely was a fan!

Finally, we should not forget that Marty was the second author of The Life of Yeast, a textbook which was fundamental to the "yeast education" of most of us.

As we hypothesized for Herman, perhaps microbiologists really do go to Heaven, and we like to imagine that Marty has already rejoined Herman in that quest for just one more new species. Maybe they have even set up shop in a replica of that lovely, messy lab that they shared for so many years in 132 Cruess Hall!

Ann Vaughan & Sandro Martini Perugia, Italy

Forthcoming Meetings

XXIIth International Conference on Yeast Genetics and Molecular Biology August 7-12, 2005, Bratislava, Slovakia

Here is a list of speakers and titles of their talks planned for each of 9 symposia organized during the meeting. Participants are reminded to book their accommodation at Academia Tours. Registration is still open at our web site (<u>http://www.yeast2005.org</u>). Although we finalized the abstract book there is still a chance to submit abstracts that will be included in a form of printed supplement as 'Late Abstracts'. We look forward to seeing you in Bratislava.

Jordan Kolarov, Chair of the XXIIth ICYGMB

Keynote lectures - Gottfried Schatz: Living with yeasts. Randy Schekman: Morphogenesis of a transport vesicle.

1. Genome stability and dynamics - Joachim Lingner: Telomerase and the mechanism of telomere length homeostasis. David Shore: Regulating telomerase at double-strand breaks and native telomeres. Virginia A.Zakian: The yeast Pif1p helicase removes telomerase from DNA ends. Raymund Wellinger: Telomerase Regulation by Yeast RNase III.

2. Evolutionary and comparative genomics - Manolis Kellis: Global views in yeast genomics: genes, regulation, evolution. Edward J. Louis: Population Genomics and Genome Evolution in Saccharomyces sensu stricto. Kenneth H. Wolfe: Rapid speciation associated with reciprocal gene loss in allopolyploid yeasts. Claude Gaillardin: Evolution in the Hemiascomycete phylum.

3. Genome, transcriptome and proteome analysis -Thomas Preiss: Gene expression and the adenylation state of the yeast transcriptome. Frederic Devaux: Transcriptome dynamics in yeast: welcome to the fourth dimension of functional genomics. Jennifer Gerton: Determinants of cohesin localization. Matthias Mann: Yeast as a model system for signaling and quantitative proteomics.

4. Chromosome segregation and replication -Tomoyuki Tanaka: Kinetochore capture and bi-orientation on the mitotic spindle. John Diffley: DNA Replication Control. Y. Watanabe: Cohesion-mediated regulation of monopolar attachment at meiosis I. Jim Haber: Multiple mechanisms of DNA repair.

5. Nuclear structure and function - Jack Greenblatt: Protein complexes and functional pathways in nuclear function. Dan Gottschling: The effects of old age on the genome. Susan Gasser: Transcription, telomeres and chromatin dynamics. Jane Mellor: Regulated transcription elongation on a chromatin template.

6. Yeast as a cognitive system: Receptors, sensors and response to the environment - Ladislav Kovac: Yeast as a cognitive system. Ian Dawes: Cellular Responses to Oxidative Stresses. Zdena Palkova: Multicellularity, advantage for long-term survival of yeast colonies. Stefan Hohmann: The response of yeast cells to osmotic stress.

7. Yeasts as pathogens - Geraldine Butler: *Candida* parapsilosis: genomic analysis and biofilm development. Dominique Sanglard: Molecular mechanisms of antifungal resistance and tolerance in *Candida albicans*. Alistair Brown: Role of transcriptional repression in *Candida albicans* pathobiology. Christophe d'Enfert: Post-genomic approaches to the study of biofilm formation by pathogenic *Candida*.

8. Yeast as a model of human diseases, ageing and apoptosis - Ulrich Brandt: The strictly aerobic yeast *Yarrowia lipolytica* as a model of human mitochondrial disorders. David Sinclair: Yeast as a model for the regulation of lifespan. Frank Madeo: New yeast homologs of mammalian apoptosis regulators. Michael Resnick: A yeast approach to human master regulatory genes, networks and master genes of diversity.

9. Organelle biogenesis, intracellular trafficking and membranes - Nikolaus Pfanner: Import and assembly of mitochondrial proteins. Takehito Shibata: Genetic diversification and unification of multicopy genes and genomes by machinery for homologous genetic recombination: Homoplasmy by Mhr1 protein of *Saccharomyces cerevisiae*. Jody Nunnari: The mechanism of mitochondrial fusion. Stephen Sturley: Conservation, divergence and convergence of transport systems for membrane lipids.

ISSY 2005 Yeast Cell Surface Orpesa (Castelló) Spain, September 28 to October 2, 2005

On behalf of the International Commission on Yeasts (ICY) we cordially invite our colleagues of the world yeast community to participate in the XXIV International Specialized Symposium on Yeasts, ISSY 2005, which will be held in Orpesa (Castelló) Spain, from September 28th to October 2nd, 2005. Scientific contributions concerning recent developments in Yeast Cell Surface research are welcome.

A brochure and a poster of this symposium are available and can be downloaded from our website:

http://www.uv.es/issy/

ISSY 2006 Systems Biology and Metabolic Engineering of Yeasts June 18-22 2006, Hanasaari, Finland

The ISSY 2006 meeting on Systems biology and metabolic engineering of yeasts will be held June 18-22, 2006, at Hanasaari, an island on the edge of Helsinki.

For further information, contact

Dr. M. Penttilä



<merja.penttila@vtt.fi>

Brief News Items Position Available - Fermentation Specialist



Fermentation Specialist / Yeast / Fuel Ethanol Location: Montréal

Description: Will report to the Vice-President R&D/QA. Will initiate and carry out fuel ethanol oriented research programs based on yeast/bacterial physiology, and coordinate technology transfer.

Requirements: PhD with 3-5 years of experience in

Contact:

Trang Dai Nguyen

yeast microbial physiology/fermentation or other appropriate scientific experience (Fermentation Engineering) with a significant record of achievement in this area. Knowledge of fermentation processes, English-French bilingualism, and supervisory experience managing staff, students/trainees and post doctoral fellows will be considered assets.

Tel. 1 514 522 2133 info@lallemand.com

PhD Student Position, Mycothèque de l'Université catholique de Louvain

A PhD position is available for 2 years starting in November 2005 with the possibility of extension. We are seeking an enthusiastic, highly motivated PhD student with a Master or Diploma degree in biological science, who will join a group working on the species delineation of yeasts. The suitable candidate will be experienced with microbiological and molecular biology techniques and should be proficient in the use of computer programs. The candidate will have a strong interest

Dr. Ing. Heide-Marie Daniel BCCM/MUCL Mycothèque de l'Université catholique de Louvain Croix du Sud 3, Bte 6 B-1348 Louvain-la-Neuve Belgium in yeast research. She/he will work in a multidisciplinary team. A sound command of the English language and basic knowledge of the French language are of advantage. The position is funded by the European Commission according to the guidelines of the Université catholique de Louvain. Applications should include a full CV and the name and contact details of two referees. The selection will be done in late October 2005. Further information can be obtained from:

Tel.: 010-4739 56 Fax: 010-4515 01 daniel@mbla.ucl.ac.be http://www.belspo.be/bccm/

Postdoctoral Position Available Molecular Mycology Research Laboratory Westmead Hospital, Sydney, Australia

A three-year postdoctoral position is available for 3 years at the Molecular Mycology Research Laboratory at Westmead Hospital, Sydney, Autralia. The research topic is: "Phylogeny as a basis for molecular identification of pathogenic fungi." The appointee will undertake research aimed at improving our understanding of the phylogeny of pathogenic fungi by indepedently undertaking sequence analysis of the rDNA gene cluster evolving genes from mainly ascomycetous yeasts, in order to contribute to the international AFTOL (Assembling the Fungal Tree of Life) project. In addition an online database containing standard PCR-fingerprinting profiles of all human pathogenic fungal species should be established. The work will utilise collections of pathogenic yeasts from the culture collection of the Molecular Mycology Research laboratory at Westmead Hospital and the Centraalbureau voor Schimmelcultures, (CBS, Utrecht, The Netherlands) as well as clinical isolates obtained from collaborating Australian microbiology labs. It is

Dr. Wieland Meyer

hoped that better knowledge of the genetic variability in clinical specimens will lead to develop a simple, reproducible, rapid and universally applicable molecular identification system, and an associated molecular reference database, for human pathogenic fungi from pure culture, tissues or body fluids. Essential criteria for this appointment include: a PhD in molecular genetics/phylogeny; skills in the areas of gene amplification from fungal DNA, phylogenetic sequence analysis, genotyping using microsatellites, computing including the use of PAUP, maximum likelihood methods, Bayesian approaches, and statistics; good verbal and written communication skills in English. Desirable criteria include training in fungal phylogeny, population genetics, publications in peer reviewed international journals; experience in fungal culturing, EMBL/GenBank database sequence submission/searches, PCR-fingerprinting, RAPD, AFLP and MLST typing. Contact:

w.meyer@usyd.edu au