

## A Newsletter for Persons Interested in Yeast

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## **Editorials**

### **Printed and Electronic Subscriptions**

A reminder of current 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). To facilitate accounting and administration, the subscription is due immediately upon receipt of the invoice that accompanies the December issue. Credit card payments can no longer be accepted because of excessive costs.

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 <lachance@uwo.ca>.

Readers who have not renewed for 2005 were sent, in April, June, and October 2005, reminder cards indicating that their subscriptions were due. Readers who have not replied were removed from the mailing list. Please encourage your colleagues who should be readers of the Yeast Newsletter to contact me for a subscription, as further reminders will not be sent.

## Yeast Photomicrographs on the Web

Linda Barnett has put 1600 photomicrographs of yeasts on the NCYC website (http://www.ncyc.co.uk/). Amongst them are nearly all the photomicrographs in Yeasts: Characteristics and Identification, 3rd edn, Barnett JA, Payne RW, Yarrow D. (2000) Cambridge University Press. These pictures (in jpeg format) are freely available for personal use. High resolution (tif) copies can be sent (in small numbers!) for individual use, without charge. There would be a charge for copies required for commercial purposes. For further details, please contact James Barnett, School of Biological Sciences, University of East Anglia, Norwich NR4 7TJ, England <j.barnett@uea.ac.uk>.

I wish all our readers a happy and scientifically rewarding New Year!

M. A. Lachance Editor

### I Culture Collection of Yeasts, Institute of Chemistry, Dúbravská cesta 9, 845 38 Bratislava, Slovakia. Communicated by Emilia Breierová <chememi@savba.sk> <u>www.chem.sk/yeast</u>

The following are abstracts of articles and lectures or posters that were published recently.

1 Miadokova E, Svidova S, Vlčkova V, Dúhova V, Nad'ová S, Rauko P, Kogan G 2006 Diverse biomodulatory effects of glucomannan from *Candida utilis*. Toxicology in Vitro 20:649–657.

Using four experimental model systems, it was demonstrated that glucomannan (GM) isolated from the cell wall of the industrial yeast *Candida utilis* revealed a broad range of protective activities. This effect depended on the nature and mode of action of the counteracting genotoxic compound as well as on the experimental model system used. In the Saccharomyces bioprotectivity assay, GM increased resistance towards ofloxacin-induced toxicity in the wild type and recombination repair-deficient yeast strains significantly enhancing survival of the cells. In the chromosomal aberration assay, GM exerted anticlastogenic effect against maleic hydrazide induced clastogenicity in *Vicia faba* L. In the DNA-topology assay, GM protected plasmid DNA from the breaks induced by Fe2+ ions,

but enhanced damage induced by bleomycin and hydrogen peroxide. In the cell-revitalization assay, it enhanced cytotoxic/cytostatic effect of teniposide applied to mouse leukemia cells. Thus, depending on the experimental model, GM acted as antimutagen, anticlastogen, DNA breaks inhibitor or inducer, and as cytotoxic/cytostatic effect enhancer. Several possible mechanisms of bioprotective action underlying the observed activities are suggested including iron chelation and free radical scavenging. The results imply that GM is a polysacharide with marked biological activities and suggest its potential biomedical application, especially in combination with other bioactive compounds.

2 Zekovič D, Radulovič M, Nastasovič A, Vrvič M.M, Jakovljevič D, Kogan G 2006 Mild Pfitzner-Moffat Oxidation of the (1-3)-β-D-Glucan from *Saccharomyces cerevisiae*. Chem Pap 60:243-248.

The structure of the cell wall glucan isolated from the industrial strain of *Saccharomyces cerevisiae* was characterized as to be composed of a linear (1-3)- $\beta$ -d-glucan chain with single  $\beta$ -gluco-pyranosyl residues attached to every ninth backbone unit by (1-6)-glycosidic linkages. Mild oxidation of this  $\beta$ -d-glucan with a dimethyl sulfoxide-acetic anhydride reagent yielded an

"oxidized" glucan with aldehyde groups introduced at C-6 and carbonyl oxygens located at C-2 and C-4 of the glucopyranosyl rings. The conversion of the oxidized glucan into the polyoxime was used to study the progress of oxidation and to establish the carbonyl groups distribution in this new reactive polysaccharide derived from baker's yeast cell wall.

3 Vlčková V, Naïová S, Dúhová V, Závodná K, Moráňová Z, Rauko P, Kogan G, Miadoková E 2006 Natural microbial polysaccharide sulphoethyl glucan as antigenotoxic and cancer preventing agent. Neoplasma 53:6.

Naturally occurring polysaccharides isolated from the yeasts are the substances with versatile intriguing biomodulatory activities. One of the novel derivatives prepared from the (1-3)-β-D-glucan isolated from the cell walls of baker's yeast *Saccharomyces cerevisiae* is sulfoethyl glucan (SEG). Its DNA-protective, antimutagenic, anticlastogenic and cytotoxic/cytostatic enhancing effect was evaluated using five eukaryotic systems. SEG showed bioprotective effect in recombination-repair-deficient strain of alga *Chlamydomonas reinhardtii* against methyl methanesulfonate-induced

genotoxicity, antimutagenic effect against ofloxacin-induced genetic changes in yeast *Saccharomyces cerevisiae* assay and anticlastogenic activity in plants *Vicia sativa* and *Vicia faba* assays against maleic hydrazide-induced clastogenicity. In the combined application with cytostatic drug vumon, SEG exerted enhancement of the drug's cytotoxic/cytostatic effect in the cell revitalization assay using mouse leukemia cells. The study sheds light on the possible mechanisms of actions and utilization of this microbial polysaccharide derivative in the cancer prevention and therapy.

# 4 Stratilová E, Dzúrová M, Breierová E, Malovíková A, Omelková J 2006 The life style of *Aureobasidium pullulans* and the multiple forms of its polygalacturonase. Biologia, Bratislava 61:257-262.

The life style of *Aureobasidium pullulans* on pectin medium and its production of extracellular polygalacturonase are closely related. Polygalact-uronases with random action pattern (EC 3.2.1.15)were formed in the first phases of cultivation, whereas exopolygalacturonases (EC 3.2.1.67 with a terminal action pattern on pectin were produced during the whole growth

of this yeast-like fungus. The production and inactivation of individual enzyme forms during cultivation were strongly dependent on the pH value of the pectin medium. Various kinds of stress can support the prolongation of the phase of endo-acting enzyme production, as well as the increase of their activity.

### 5 Maceková D, Farkaš V, Kishida E, Takeo K 2006 Ecto-glycanases and metabolic stability of the capsule in *Cryptococcus neoformans*. J Basic Microbiol 46:470-479.

We have identified a number of ecto-glycanases (glycosylhydrolases) associated with the capsule and/or the cell wall of *Cryptococcus neoformans*. The enzyme activities detected included  $\alpha$ -mannosidase,  $\alpha$ - and  $\beta$ -glucosidase,  $\alpha$ - and  $\beta$ -glactosidase,  $\beta$ -xylosidase,  $\beta$ -glucuronidase, and endo- $\beta$ -1,3-glucanase. Small portions of the enzymes were also secreted into the growth medium. Cell-wall associated endo- $\beta$ -1,3-glucanases exhibited highest activity in the acidic range between pH 2.5 and 5.0. The products of laminarin hydrolysis by the enzymes located on the cell surface were glucose and  $\beta$ -1,3-linked glucooligosaccharides. The same products were released from isolated cell walls incubated in the buffer. Endo- $\beta$ -1,3-glucanase activity extracted from the cell surfaces by mild sonication

consiste of six isoforms separable by isoelectric focusing. In spide of the presence of the whole array of glycanase activities on the cell surfaces, capsular polysaccharides released from *C. neoformans* cells into the growth medium were practically metabolically stable. From the defined polysaccharides tested, only laminarin ( $\beta$ -1,3-glucan) and to some extent also mixed-linkage  $\beta$ -1,3/ $\beta$ -1,4-glucan and/or 4-O-methyl-D-glucurono-D-xylan were able to support the yeast growth. The activities of majority of identified ecto-glycanases were low when the yeast was grown on glucose but were considerably elevated when the cells were grown on glycerol indicating that their synthesis is regulated by catabolite repression.

### 6 Mazáň M, Mazáňová K, Farkas V 2006 Fungal cell wall - challenge for the search of new antimycotics. Chem Listy 100:433-439.

The microorganism cell wall is a solid but dynamic structure, which is essential for the life of cells in a changing environment. Chemical composition of the fungal cell wall differs from those of surface structures of mammalian cells and plant cell walls; hence, the fungal cell wall is an ideal target organelle for a new generation of more efficient and less toxic antimycotics. For that reason it is important to know the composition of fungal cell wall, how its components are synthesized and how their biosynthesis is regulated. The review summarizes the present state of knowledge of the yeast cell wall and its biosynthesis.

7 Naďová S, Vlčková V, Dúhová V, Pražmáriová E, Špinerová L, Kogan G, Rauko P, Miadoková E 2006 The evaluation of biomodulatory effects of natural compounds. Lecture: Aktuální Problematika Genetické Toxikologie 29. pracovní dny České a slovenské společnosti pro mutagenezi zevním prostøedím pøi Československé biologické společnosti Brno, ČR, 2.–4. 5. 2006, NCO NZO, Zbornik (J. Topinka, Ed.) ISBN 80-7013-438-0, pp. 51-54.

The need of finding the new compounds with specific biomodulatory abilities increases clearly show their marked immunological properties such as antioxidant, anticancer or antimutagenic activities or their other non-specific stimulations of the host immune system. The main aim of this work was to evaluate the potential biomodulatory abilities of natural polysaccharide carboxymethyl glucan (CMG) and the plant extract of *Cynara cardunculus* L. (ECC), rich on flavonoids apigenin and luteolin and their glycosides, by using the following genetic model systems: bacteria *Salmonella typhimurium*, a unicellular green alga *Chlamydomonas reinhardtii*, yeast *Saccharomyces cerevisiae*, plant *Vicia sativa* L, murine leukaemia L1210 cells and pBR322 plasmid DNA from *Escherichia coli*.

- 8 S Naďová, E Pražmáriová, V Vlčková, V Dúhová, G Kogan, P Rauko, E Miadoková 2006 Carboxymethyl glucan-polysaccharide with antigenotoxic potential. Poster: Aktuální Problematika Genetické Toxikologie 29. pracovní dny České a slovenské společnosti pro mutagenezi zevním prostøedím pøi Československé biologické společnosti Brno, ČR, 2.–4. 5. 2006, NCO NZO, Zbornik, pp. 97-98
- 9 TA Korolenko, TV Alexeenko, SYa Zhanaeva, AA Venediktova, G Kogan, NN Besednova, TA Kuznetzova, TN Zviagintzeva, VI Kaledin 2006 Effect of immunomodulator biological response modifiers on murine tumors and metastases. Poster: 4th Conference on Experimental and Translational Oncology Kranjska gora, Slovenia, March 22-26, 2006. Book of Abstracts, p. 88.
- 10 Miadoková E, Naïová S, Pražmáriová E, Dúhová V, Vlčková V, Kogan G, Rauko P 2006 The role of natural polysacharide as agent with antimutagenic/anticarcinogenic activity. Poster P-098, From Genes to Molecular Epidemiology, 36<sup>th</sup> Annual Meeting of the European Environmental Mutagen Society, Prague, Czech Republic, July 2-6, 2006. Book of Abstracts, p. 190.

- 11 G Kogan, E Miadoková, D Slameňová, V Vlčková, M Babincová, P Rauko 2006 Antioxidant, antigenotoxic, and immunomodulating properties of yeast cell wall polysaccharides. Lecture, 20<sup>th</sup> Biochemical Congress, Piešžany, September 12-16, 2006-09-18 Book of Proceedings, p. 100, ISBN 80-969532-6-5.
- 12 G Kogan, D Slameňová, P Rauko, A Staško, M Babincová, TA Korolenko 2006 Could Fungal Polysaccharides Fight Cancer? Lecture, 2<sup>nd</sup> Baltic Meeting on Microbial Carbohydrates, Rostock, Germany, October 4-8, 2006. Book of Abstracts, p. 57.
- 13 M Pajtinka, Z Sejáková, G Kogan, K Dercová 2006 Sorption capacities of cell wall glucan isolated from Saccharomyces cerevisiae towards PCP. Lecture, 2<sup>nd</sup> Baltic Meeting on Microbial Carbohydrates, Rostock, Germany, October 4-8, 2006. Book of Abstracts, p. 56.
- 14 E Miadoková, S Nadová, V Vlčková, V Dúhová, E Pražmáriová, P Rauko, L Čipák, G Kogan 2006 Antigenotoxic aspects of natural compounds actions. Short lecture, Molecular and phzsiological effects of bioactive food compounds, COST 926/927 Conference, Vienna, Austria, October 11-14, 2006. Book of Abstracts, p. 178.
- 15 Čertík M, Breierová E, Márová I, Masrnová S. Physiological regulation of microbial pigment production. Lecture: 97<sup>th</sup> AOCS Annual Meeting & Expo, America's Center St.Louis, Missouri, April 30-May 3, 2006. Book of Abstracts, p.17-18.
- II Department of Microbial, Biochemical & Food Biotechnology / UNESCO MIRCEN, University of the Free State, P.O. Box 339, 9300 Bloemfontein, South Africa. Communicated by James du Preez <dpreezjc.sci@mail.uovs.ac.za> www.uovs.ac.za/biotech

The following articles from our department have recently appeared or are in press.

- 1 Bareetseng AS, Kock, JLF, Pohl CH, Pretorius EE, Strauss CJ, Botes PJ, Van Wyk PWJ, Nigam S 2006 Mapping the distribution of 3-hydroxy oxylipins in the ascomycetous yeast *Saturnispora saitoi*. Syst Appl Microbiol 29:446-449.
- 2 Leeuw NJ, Kock JLF, Pohl CH, Bareetseng AS, Sebolai OM, Joseph M, Strauss CJ, Botes PJ, Van Wyk PWJ, Nigam S 2006 Oxylipin covered ascospores of *Eremothecium coryli*. Antonie van Leeuwenhoek 89:91-97.
- 3 Strauss CJ, Van Wyk PWJ, Lodolo EJ, Botes PJ, Pohl CH, Nigam S, Kock JLF 2006 Oxylipin associated co-flocculation in yeasts. JIB 112:66-71.
- 4 Leeuw NJ, Swart CW, Ncango DM, Pohl CH, Sebolai OM, Strauss CJ, Botes PJ, van Wyk PWJ, Nigam S, Kock JLF 2006 Acetylsalicylic acid as antifungal in *Eremothecium* and other yeasts. Antonie van Leeuwenhoek (In press).
- 5 Kock JLF, Strauss CJ, Pohl CH, Van Wyk PWJ, Botes PJ 2006 Yeast Biomechanics. Proceedings: III European Conference on Computational Mechanics Solids, Structures and Coupled Problems in Engineering. Lisbon, Portugal, 5-8 June 2006, Eds. C.A. Mota Soares et al. p. 725. ISBN-101-4020-4994-3 (HB) & ISBN-13 978-1-4020-4994-1 (HB). Springer, The Netherlands.
- 6 Ncango DM, Pohl CH, Sebolai OM, Botes PJ, Strauss CJ, Joseph M, van Wyk PWJ, Nigam S, Kock JLF 2006 Oxylipin-coated hat-shaped ascospores of *Ascoidea corymbosa*. Can J Microbiol (In press).
- 7 Van Heerden A, Van Wyk PWJ, Botes PJ, Pohl CH, Strauss CJ, Nigam S, Kock JLF 2006 The release of elongated, sheathed ascospores from bottle-shaped asci in *Dipodascus geniculatus*. FEMS Yeast Res (In press).

8 Pohl CH, Van Wyk PWJ, Kock JLF, Albertyn J 2006 *Cryptococcus anemochorus* sp. nov. a new anamorphic basidiomycetous yeast isolated from the atmosphere in central South Africa. Int J Syst Evol Microbiol 56:2703-2706.

*Cryptococcus anemochorus* was isolated from the atmosphere in central South Africa. Sequence analysis of the D1/D2 domain and internal transcribed spacer region indicate that *Cryptococcus anemochorus* is a novel species within the *Cryptococcus laurentii* complex. Phylogenetic analyses based on the D1/D2 domain revealed that it occupies a relatively isolated position within this complex with *Papiliotrema bandonii*, *Cryptococcus perniciosus*, *Cryptococcus nemorosus* and *Cryptococcus* sp. CBS 8363 being the closest relatives. However,

this species could be distinguished from related species by standard physiological tests including the inability to assimilate rhamnose, meth-a-glucoside, salicin, lactose, erythritol, ribitol, xylitol, citrate and ethanol. In addition no extra cellular starch production was observed and the isolate was able to grow in the absence of additional vitamins. On the basis of these results we propose *Cryptococcus anemochorus* sp. nov. (CBS  $10258^{T} = NRRL Y-27920^{T}$ ).

1. Shiningavamwe A, Obiero G, Albertyn J, Nicaud JM, Smit MS 2006 Heterologous expression of the benzoate *para*-hydroxylase encoding gene (*CYP53B1*) from *Rhodotorula minuta* by *Yarrowia lipolytica*. Appl Microbiol Biotechnol 72:323-329.

There is currently an increasing number of cytochrome P450 (CYP450) monooxygenase encoding genes becoming available from various genome-sequencing projects. These enzymes require association with cytochrome P450 reductase (CPR) to achieve optimal activities. In this study, the CYP53B1 gene, which encodes a benzoate parahydroxylase, was successfully cloned from *Rhodotorula minuta* and overexpressed in *Yarrowia lipolytica* E150. Multiple copies of the CYP53B1 cDNA were cloned under the POX2 promoter, while the *Y. lipolytica* CPR was cloned under the isocitrate lyase promoter. Whole cell biotransformation of benzoic acid to parahydroxylase activity of the recombinant *Y. lipolytica* UOFS

Y-2366. Different induction conditions were tested in shake flask cultures. The highest concentration of pHBA produced by UOFS Y-2366 was 1.6 g l<sup>-1</sup> after 200 h when stearic acid was repeatedly added to the media. *R. minuta* accumulated up to 1.8 g l-1 of pHBA within only 24 h. Thus, the specific hydroxylase activity of *Y. lipolytica* UOFS Y-2366 [approximately 0.07 U (g dry wt.)<sup>-1</sup>] was about 30 times lower than the specific hydroxylase activity of *R. minuta* [2.62 U (g dry wt.)-1]. However, the hydroxylation activity obtained with *Y. lipolytica* was one of the highest hydroxylation activities thus far reported for whole cell biotransformation studies carried out with yeasts expressing foreign CYP450s.

## III Rosenstiel School of Marine and Atmospheric Science, University of Miami, 4600 Rickenbacker Causeway, Miami, Florida 33149, USA. Communicated by J.W. Fell <a href="mailto:spinoremailto:fell@rsmas.miami.edu">fell@rsmas.miami.edu</a>.

Recent publications from our laboratory.

- 1 Fell JW, Scorzetti G, Connell L, Craig S 2006 Biodiversity of micro-eukaryotes in Antarctic dry valley soils with < 5% soil moisture. Soil Biochem 38:3107-3115.
- 2 Adams BJ, Bardgett RD, Ayres E, Wall DH, Aislabie J, Bamforth S, Bargagli R, Cary C, Cavacini P, Connell L, Convey P, Fell JW, Frati F, Hogg ID, Newsham KK, O'Donnell A, Russell N, Seppelt RD, Stevens MI 2006 Diversity and Distribution of Victoria Land Biota. Soil Biol Biochem 55:3003-3018.
- 3 Diaz MR, Boekhout T, Bovers M, Cabanes FJ, Fell JW 2006 Microcoding and flow cytometry as a highthroughput fungal identification system for *Malassezia* species. J Med Microbiol. 55:1197-1209.

## IV Russian Collection of Microorganisms (VKM), Institute for Biochemistry and Physiology of Microorganisms, Pushchino, 142290, Russia. Communicated by WI Golubev <wig@ibpm.pushchino.ru>.

Recent publications.

1 Golubev WI, Pfeiffer I, Golubeva EW 2006 Mycocin production in *Pseudozyma tsukubaensis*. Mycopathol 162:313-316.

Killer activity expressed at pH values ranging from 3.5 to 6.0 was found in the ustilaginaceous yeast-like species, *Pseudozyma tsukubaensis*. Its killer phenotype was incurable, and extrachromosomal genetic elements were not detected. The

toxin excreted with a molecular mass above 15 kDa is fungicidal, resistant to proteolytic cleavage, thermolabile and active only against fungi within the Ustilaginomycetes (the orders Microstromatales and Ustilaginales).

2 Golubev W, Sugita T, Golubev N 2007 Ustilaginomycetous yeast, *Pseudozyma graminicola* sp. nov., isolated from the leaves of pasture plants. Mycoscience (accepted).

Two strains belonging to a novel anamorphic species, *Pseudozyma graminicola*, were isolated from the leaves of herbaceous plants in Moscow region (Russia). This species was genetically distinct from all known *Pseudozyma* species, based on sequence divergence in the D1/D2 domains of the

large-subunit rDNA and the ITS region. It is related phylogenetically to species of the genus *Sporisorium* (Ustilaginaceae, Ustilaginales). Physiological characteristics distinguishing a novel species from the other species of the genus *Pseudozyma* are presented.

## 3 Golubev WI, Sampaio JP, Golubeva EW 2006 *Cryptococcus stepposus*, a new filobasidiaceous yeast species found in the Prioksko-terrasny biosphere reserve in Russia. Mycol. Res. 110:957-961.

Mycocinotyping of cryptococci from the Prioksko-terrasny reserve has revealed three strains that have a unique mycocin-sensitive profile. Sequencing results of the D1/D2 domain of the 26S rDNA and the complete ITS region placed them in the Floriforme clade of the Filobasidiales lineage (Hymenomycetes). the three strains had identical sequences,

which differed from those of known *Filobasidium* and *Cryptococcus* species. A novel species named *Cryptococcus stepposus* (type strain VKM Y-2918) is proposed to accommodate these isolates. Physiological characteristics distiguishing the novel species from other *Cryptococcus* species in the Floriforme clade are presented.

4 Kulakovskaya EV, Golubev WI, Kulaev IS 2006 Extracellular antifungal glycolipids of the yeast Cryptococcus humicola. Doklady Akad Nauk 410, N3 (in press).

All five strains of *Cryptococcus humicola* (including type strains of this species and *Nadsonia slovaca*) secreted cellobiose lipids that were active against many fungi under acidic conditions. All cells of basidiomycetous yeasts (*Cr. terreus, Filobasidiella neoformans*) were killed at cellobiose lipid

concentrations of 0.04-0.08 mg/ml whereas ascomycetous ones (*Candida albicans, C. glabrata, C. viswanathii, Clavispora lusitaniae, Saccharomyces cerevisiae*) were killed at concentration of 0.5 mg/ml.

### V Institute of Fermentation Technology and Microbiology, Technical University of Lodz, 90-924 Lodz, Wolczanska 171/173, Poland. Communicated by Dorota Kregiel <dkregiel@p.lodz.pl>.

The following article was recently published.

1 Berłowska J, Kregiel D, Klimek L, Orzeszyna B, Ambroziak W 2006 Novel yeast cell dehydrogenase activity assay *in situ*. Polish J Microbiol 2:127-131.

The aim of this research was to develop a suitable method of succinate dehydrogenase activity assay *in situ* for different industrial yeast strains. For this purpose different compounds: EDTA, Triton X-100, sodium deoxycholate, digitonin, nystatin and  $\beta$ -mercaptoethanol were used. The permeabilization process was controlled microscopically by primuline staining. Enzyme assay was conducted in whole yeast cells with Na-succinate as substrate, phenazine methosulfate (PMS) as electron carrier and in the presence one of two different tetrazolium salts: tetrazolium blue chloride (BT) or cyanoditolyl tetrazolium chloride (CTC) reduced during the assay. In comparabile studies of yeast vitality the amount of intracellular ATP was determined according to luciferin/luciferase method. During the succinate dehydrogenase assay in intact yeast cells without permeabilization, BT formazans were partially visualized in the cells, but CTC formazans appeared to be totally extracellular or associated with the plasma membrane. Under these conditions there was no linear relationship between formazan color intensity signal and yeast cell density. From all chemical compounds tested, only digitonin was effective in membrane permeabilization without negative influence on cell morphology. Furthermore, with digitonin-treated cells a linear relationship between formazan color intensity signal and yeast cell number was noticed. Significant decreasing of succinate dehydrogenase activity and ATP content were observed during aging of the tested yeast strains.

Posters presented at the International Specialized Symposium on Yeasts (ISSY 2006), June 18-21, Hanasaari, Espoo, Finland.

2 Berłowska J, Krêgiel D, Ambroziak W 2006 The pyruvate decarboxylase activity assay of yeast in situ.

Cytoplasmic pyruvate decarboxylase EC 4.1.1.1 (PDC), is one of the key yeast enzymes which allows precise and recurrent description of fermentative metabolism. PDC is the first enzyme of glycolytic pathway which, under anaerobic conditions, leads to nonoxidative decarboxylation of pyruvate with acetaldehyde as end product However, this enzyme is also capable to condensate of acetaldehyde and pyruvate and to form acetoine. There is also a known fact, that some of yeast strains could produce acetate from accumulated acetaldehyde. Therefore, in the case of enzymatic reactions *in situ*, control of all products of these metabolic pathways, in which PDC could participate, is necessary. The aim of this research was to develop a suitable method of yeast pyruvate decarboxylase activity assay *in situ* and to compare enzyme activity of yeast cells in different physiological states. Yeast *Saccharomyces cerevisiae* Bc16a - distillery strain was grown under aerated conditions on an orbital shaker in such nutrient media as YPM and wort broth with 1% and 12% maltose respectively and in fermentative medium with 12% maltose. Enzymatic assay was conducted in cell suspension with digotonin, as permabilisation agent, and sodium pyruvate, as a substrate, at temperature 30°C. PDC metabolites - acetaldehyde, ethanol, acetoine and acetate were detected using GC technique with Headspace Autosampler. Different parameters: type and concentration of substrate, minimal effective concentration of digitonin, cell density, reaction time and effect of pyrazole, as alcohol dehydrogenase inhibitor, were

checked in order to optimize PDC assay *in situ*. In the concentration range of yeast cells from  $1 \times 10^7$  to  $1 \times 10^8$ /ml the linear correlation was noticed between produced acetaldehyde and cell density. For the given conditions, in the presence of 0,05 M pyruvate and 0,05 % digitonin, the enzymatic reaction was linear up to 20. minutes of assay. During incubation there was no formation of ethanol and therefore pyrazole wasn't necessary for the assay. PDC activity in *S. cerevisiae* cells depends on physiological state of cells and culture conditions (oxygen availability, sugar concentration). The maximal PDC activity of yeast cells was observed on the 3<sup>rd</sup> day of fermentation process, whereas, on 7<sup>th</sup> day of aerobic cultivation, PDC activity was not seen.

#### 3 Kregiel D, Berłowska J, Ambroziak W 2006 Adhesion of selected yeast strains on solid type of carriers.

Cell adhesion is the fundamental phenomenon that govern and describes bioengineering processes that employ cell immobilization with focusing on different biotechnological applications, including beer and biofuel production. The microtopography of the contact surface, physiological state of cells, their phase of growth and nutrient availability are important in determining the adhesion process. Specific objective of our research was to study how immobilisation conditions: type of culture medium, cell density, physiological state of yeasts and incubation time affected cells attachment to the different solid carriers - three types of hydroxylapatite and chamotte tablets. The industrial brewery and distillery strains Saccharomyces cerevisiae and unconventional amylolytic strain Debaryomyces occidentalis from different physiological states were used in this study. Microbial adhesion was conducted in nutrient and minimal culture media. In all the experiments, for the estimation of the number of adhered cells and effectiveness of immobilization, microscopic method and methylene blue staining were applied and, in some cases, additionally, DAPI fluorymetric method was

also employed. The immobilization of selected yeasts on hydroxylapatie carriers was weak. However, when incubation of cells was conducted under starvation condition, the scale of observed cell immobilization after 24 hours was higher, especially in the case of D. occidentalis strain. The significant differences between cell density and the rate of adhesion were not observed for all tested yeast cells. Therefore we postulate that adhesion on hydroxylapatite carriers has only reversible character. The better results we observed in the case of chamotte - the number of immobilized cells was about  $10^{6}$ - $10^{7}$  cell per carrier and cell adhesion was stable during 72 hours of fermentation. However, formation of three-dimensional cell structure and microcolonies were not observed. Therefore we can conclude, that the optimal adhesion conditions should be estimated individually for each yeast strain, solid carrier and fermentation process. Determination of the most preferable conditions of adhesion will allow for efficient application of immobilized yeast cells for beer production and conversion of starch to ethanol.

# 4 Kregiel D, Dziedziczak K, Ambroziak W 2006 Characteristic of Crabtree-negative yeast *Debaryomyces occidentalis* encapsulated in foamed alginate beads.

Immobilization of whole cells by entrapment method is a simple technique widely used in research and industrial applications. Such biotechnological approach has many advantages due to better operational stability and higher efficiency of biocatalysis. Although starch offers a high-yielding ethanol source, yeast Saccharomyces cerevisiae with superior ethanol fermentation capability is unable to convert starch into ethanol due to lack of complex of amylolytic enzymes. Contrary to its abilities, unconventional yeast Debaryomyces occidentalis, described by Ingledev as a "super yeast", secretes both  $\alpha$ -amylase and glucoamylase, which together result in a complete degradation of starch into fermentable glucose. Because of these enzymes irreversible inactivation at 60°C and pH optimum between 5 to 6, they are ideally suited for commercial bioethanol or low ethanol beer production. However, the barrier in wide application of *D. occidentalis* in continuous fermentation processes is seen in fact that this respiratory, Crabtree-negative yeast requires a growth-limiting supply of oxygen to secrete amylases and to conduct alcoholic fermentation. In the present work was developed new, unique method of yeast immobilization and its utilization in specific biotechnological processes. The alginate multichamber cores with yeast cells were formed in traditional way from foamed basic solutions. The research was focused on the determination of growth patterns, cell viability and metabolism of respiratory yeast cells encapsulated inside alginate cores. The application of the novel immobilization technique for D. occidentalis resulted in a high viability of entrapped cells. The initial cell concentration in the core was 10<sup>4</sup> cfu and after period of adaptation during cultivation of the cores in the growth medium increased to  $10^7$  cfu. In the multichamber beads the further colonization with the formation of yeast microcolonies was observed. The amylolytic activity and ethanol production of free and immobilized cells D. occidentalis in starch media were comparable, but GC analysis of fermentation products showed different metabolic patterns.

# 5 Dziedziczak K, Kregiel D, Kordialik-Bogacka E, Ambroziak W 2006 The metabolic profiles of immobilized brewery yeasts.

Immobilization of microbial cells is increasingly applied in biotechnological processes. The main benefits can be seen in improved quality of biotechnologies, linked with such traditional fermentation processes as production of beer and further in open technological prospects to novel biotechnologies. Brewing industry is deeply interested, for economical reason, in brewery veast cells immobilization and its application to continuous processes. It is expected to see in immobilized cell technology the advantage of the reducing time of processes without reduction of product quality. The use of immobilized yeasts may be interesting alternative for conventional processes with increasing productivity of cells loading in bioreactors. The leading idea of this study was to use method of yeast cells entrapment inside special foamed alginate cores with internal multichambers and to investigate of cells growth in the state of immobilization. We were also interested in evaluation of their ability to conduct ethanol fermentation with specific metabolite profiles, important for proper taste and flavour. Different Saccharomyces cerevisiae

brewery, bottom-fermenting yeast strains were used. The primary fermentations were conducted in bath cultures in 12°P in malt wort at temperature 10°C during 7 days with free and immobilized yeast strains. The analysis of carbohydrates was carried out by HPLC - high-performance liquid chromatography. The flavor and aroma compounds (ethanol, higher alcohols and esters) was controlled by GC - gas chromatography analysis with Headspace Autosampler. The results of studies have shown high ethanol production by entrapped yeast cells and proper profile of green beer flavor. The production of higher alcohols and esters by free and immobilized cells was comparable, but amount of acetaldehyde produced by immobilized yeast was significantly decreased. For each tested brewery yeast strain a specific metabolic profiles were seen. Sequential passages of encapsulated yeast cells into fresh fermentation media have shown the significant reduction of time required for primary fermentation.

Poster presented at the 2<sup>nd</sup> Congress of European Microbiologists, July 4-8, Madrid, Spain.

# 6 Kregiel D, Dziedziczak K, Ambroziak W 2006 Fermentation profiles of free and immobilized yeast *Debaryomyces occidentalis* in starch medium.

Yeast metabolism is widely exploited in different biotechnological processes. The term "yeast" has in some contexts been used as a synonym to the species name Saccharomyces cerevisiae. However, the properties of S. cerevisiae don't represent all yeasts. Among the approximately 800 recognized yeast species, only few ones are used commercially. The metabolic pathways of the central carbon metabolism are generally the same between different yeast species, however, the regulation of fermentation and respiration differ substantially. Yeast Debaryomyces occidentalis is the unconventional yeast that has attracted great attention due to its capacity to grow on unusual carbon sources. D. occidentalis produces different extracellular enzymes and this yeast can completely hydrolyze soluble starch and its derivatives. However, in this respirative yeast, oxygen limitation induced both starch hydrolysis and alcoholic fermentation (Kluyver effect). The aim of this work was to study the influence of encapsulation of D. occidentalis cells in foamed alginate gels on the growth patterns and metabolic profiles in starch media. The alginate cores with yeast cells were formed from foamed basic solutions.

Yeast cell number was determined by plate count method after dissolving cores. Fermentation profiles (sugars, ethanol and acetaldehyde) were determined chromatographically (GC and HPLC). The application of the novel immobilization technique for respiratory yeast cells resulted in a high viability of encapsulated cells. The initial cell concentration in the bead was 10<sup>4</sup> cfu/core and after a prolonged period of adaptation phase during cultivation of the beads in the growth medium increased to  $1-2 \times 10^7$  cfu/core. The amylolytic activity and ethanol production of free and immobilized cells D. occidentalis in starch media were comparable, but chromatography analysis of other fermentation products showed different metabolic patterns, especially for acetaldehyde and ethyl acetate production. However first passage of encapsulated cells into fresh fermentation media led to similarity in profiles. This fact confirms adaptation of D. occidentalis cells to the special enological environment in alginate gels and possibility of using this yeast in mono- and mixed immobilized populations for bioconversion starch to ethanol.

These works were partially supported by UE 6PR Grant NMP3-CT-2003- 504937 PERCERAMICS and KBN Grant 2 P06T 081 29.

VI Laboratorio de Microbiología Aplicada y Biotecnología (Applied Microbiology and Biotechnology Laboratory), Centro Regional Universitario Bariloche, Universidad Nacional del Comahue, Quintral 1250, (8400), Bariloche, Argentina. Communicated by Diego Libkind <libkind@crub.uncoma.edu.ar>.

Recent Publications.

- 1 Pérez P, Libkind D, Diéguez MC, Summerer M, Sonntag B, Sommaruga R, van Broock M, Zagarese HE 2006 Mycosporines from freshwater yeasts: a trophic cul-de-sac? Photochem Photobiol Sci 5:25-30. The abstract was given in the Spring issue.
- 2 Libkind D & van Broock MR 2006 Biomass and carotenoid pigments production by Patagonian native yeasts. World J Microbiol Biotechnol 22:687-692.

New yeast isolates from unexplored Patagonian habitats were studied for the production of biomass and carotenoids as the first step towards the selection of hyper-producing strains and the design of a process optimization approach. Patagonian yeast isolates considered as potential biomass and carotenoid sources were studied using ammonium sulphate and urea as nitrogen sources in semi-synthetic medium (MMS), and agro-industrial byproducts (cane molasses, corn syrup, raw malt extract) as carbon sources. Maximum pigment production (300 ig  $g^{-1}$ ) was achieved by *Rhodotorula mucilaginosa* CRUB 0195 and by novel species *Cryptococcus* sp. CRUB 1046. â-carotene, torulene and torularhodin were the major carotenoids found.

3 Libkind D, Diéguez M, Moliné M, Pérez P, Zagarese H, van Broock M 2006 Occurrence of photoprotective compounds in yeasts from freshwater ecosystems of northwestern Patagonia (Argentina). Photochem Photobiol 82:972-980.

In this paper we present the results of research on the occurrence, induction and role of photoprotective compounds (PPCs) present in native aquatic yeasts from freshwater Patagonian ecosystems. We focus on the effect of UV radiation (UVR) as a factor that controls the level of photoprotection of yeasts, and explore its potential significance in shaping yeast distributional patterns. The research presented here combines field surveys and laboratory work, including the isolation and culture of native yeasts strains, and laboratory assays under different radiation conditions. The results obtained suggest that yeasts are common dwellers of oligotrophic Patagonian water bodies, and provide the first evidence of the distribution of PPC (carotenoid and mycosporine)–producing yeasts in temperate freshwaters. A greater proportion of carotenogenic yeasts were

observed in high-elevation lakes. The yeast strains isolated from these environments were found to produce higher amounts of mycosporines (MYCs), and to present higher tolerance to UVB exposure than those from piedmont lakes. Patagonian yeasts have only one type of MYC, mycosporine-glutaminol-glucoside (mycglu-glu), which seems common to all other yeasts. By analyzing the production of myc-glu-glu in a large number of yeasts belonging to different taxonomic groups, we propose that this compound may have potential use as a chemotaxonomic marker in yeast systematics. Collectively, our work reveals that in Patagonian freshwater yeasts there is an apparent relationship between the ability to produce PPCs, their tolerance to UV exposure and their success in colonizing habitats highly exposed to UVR.

Publications in Press.

# 4 Libkind D, Ruffini A, van Broock M, Alves L, Sampaio JP - Biogeography, host-specificity and molecular phylogeny of *Phaffia rhodozyma* and its sexual state *Xanthophyllomyces dendrorhous*. Appl Environ Microbiol.

*Phaffia rhodozyma* (sexual state: *Xanthophyllomyces dendrorhous*) is a basidiomycetous yeast that has been found in tree exudates in the northern hemisphere at high altitudes and latitudes. This yeast produces astaxanthin, a carotenoid pigment with biotechnological importance because it is used in aquaculture for fish pigmentation. We isolated *X. dendrorhous* from the southern hemisphere (Patagonia, Argentina), where it was associated with fruiting bodies of *Cyttaria hariotii*, an ascomycetous parasite of *Nothofagus* trees. We compared ITS-based phylogenies of *P. rhodozyma* with that of its tree host (Betulaceae, Corneaceae, Fagaceae and Nothofagaceae) and found them to be generally concordant, suggesting that different yeast lineages colonize different trees and providing an

explanation for the phylogenetic distance observed between the type strains of *P. rhodozyma* and *X. dendrorhous*. We hypothesize that the association of *Xanthophyllomyces* with *Cyttaria* derives from a previous association of the yeast with *Nothofagus*, and the sister relationship between Nothofagaceae and Betulaceae plus Fagaceae correlates with the phylogeny of *X. dendrorhous* strains originating from these three plant families. The two most basal strains of *X. dendrorhous* are those isolated from *Cornus*, an ancestral genus in the phylogenetic analysis of the host trees. Thus, we question previous conclusions that *P. rhodozyma* and *X. dendrorhous* represent different species since the polymorphisms detected in the ITS and IGS sequences can be attributed to intraspecific variation associated with host-

5 de García V, Brizzio S, Libkind D, Buzzini P, van Broock M 2006 Biodiversity of cold-adapted yeasts from runoff glacial rivers in Patagonia, Argentina. FEMS Microbiol Ecol.

The occurrence of culturable yeasts in glacial meltwater from the Frías, Castaño Overo and Río Manso glaciers, located on Mount Tronador in the Nahuel Huapi National Park (Northwestern Patagonia, Argentina), is presented. Subsurface water samples were filtered for colony counting and yeast isolation. The total yeast count ranged between 6 and 360 CFU  $L^{-1}$ . Physiologic and molecular methods were employed to identify 86 yeast isolates. In agreement with yeast diversity data from studies for Antarctic and Alpine glaciers, the genera *Cryptococcus, Leucosporidiella, Dioszegia, Rhodotorula*, *Rhodosporidium*, *Mrakia*, *Sporobolomyces*, *Udeniomyces* and *Candida* were found. *Cryptococcus* and *Leucosporidiella* accounted for 50% and 20% of the total number of strains, respectively. Among 21 identified yeast species, *Cryptococcus* sp. 1 and *Leucosporidiella fragaria* were the most frequent. The typically psychrophilic *Mrakia* yeast strain and three new yeast species, yet to be described, were also isolated. All yeast strains were able to grow at 5, 10, and 15 °C. Among yeast strains expressing extracellular enzymatic activity, higher proteolytic and lipolytic activities were obtained at 4 °C than at 20 °C.

Publications submitted or in preparation.

- 6 Brizzio S, Turchetti B, de García V, Libkind D, Buzzini P, Gasparetti C, van Broock M Extracellular enzymatic activities (EEA) in basidiomycetous yeasts isolated from glacial and subglacial waters of northwest Patagonia (Argentina). Can J Microbiol.
- 7 Moliné M, Libkind D, Diéguez MC, van Broock M Photo-protective role of carotenoid pigments in yeasts: experimental study contrasting naturally occurring pigmented and albino strains. Photochem Photobiol Sci.
- 8 Libkind D, Gadanho M, van Broock M, Sampaio JP Intraspecific variability in *Rhodotorula mucilaginosa* yeast strains from Patagonia, Argentina.
- 9 Russo G, Libkind D, Sampaio JP, van Broock Yeast biodiversity at the Volcanic acidic environment of the Lake Caviahue and Rio Agrio (Patagonia, Argentina).
- 10 Libkind D, Arts M, van Broock M Fatty acid composition of carotenogenic yeasts.
- 11 Diego Libkind Frati September 25 2006 PhD in Biochemistry, Universidad Nacional de Tucumán, Argentina.

This PhD thesis was focused on the study of pigmented veast diversity in aquatic environments of northwestern Patagonia. These are unexplored habitats with respect to their microbiology. In addition, it aimed to assess the biotechnological potentialities of the isolated yeasts in relation to the production of carotenoid pigments, UV absorbing compounds, extracellular enzymes and lipids. Fifteen waterbodies were studied within the Nahuel Huapi National Park (NHNP), where yeast counts were variable (< 2 - 1.668 UFC L<sup>-1</sup>). Increasing yeast values were observed with the cost proximity and with anthropic influence. All the aquatic environments surveyed had pigmented yeasts in proportions that increased along with the coast's distance and water transparency. 201 yeast strains were isolated and identified using conventional and molecular techniques (Micro/Minisatellite-Primed PCR fingerprinting and rADN sequencing). They were classified in 8 genera and 25 species belonging to 4 taxonomic groups: Sporidiobolales, Erythrobasidium clade, Cystofilobasidiales and Tremellales. Rhodotorula mucilaginosa was the most abundant species and the most widely distributed. Six novel yeast species were found, 2 of which were described as Sporobolomyces patagonicus and Sporidiobolus longiusculus. The existence of *Xanthophyllomyces dendrorhous* in natural habitats of the southern hemisphere was described for the first time. Additional studies revealed that this species was associated to sugary stromata of *Cyttaria hariotii* fungi, an obligate parasite of *Nothofagus* spp. trees, and that patagonian strains were genetically distinct probably due to their geographic isolation and specificity of habitat.

Carotenoid pigment synthesis was stimulated by photosintetically active radiation (PAR), in particular in those strains with low levels of constitutive carotenoids. The synthesis of UV absorbing compounds such as mycosporines is described for the first time in yeasts. PAR exposition induces the accumulation of this metabolite. A higher induction was observed in cultures exposed to PAR plus UV radiation. Only those species belonging to the *Erythrobasidium* clade and the Tremellales order were mycosporinogenic. These species produce a single compound identified as micosporine-glutaminolglucoside (MGG), with a maximum absorbance at 310 nm, which could be useful as chemo-taxonomic marker in yeast systematic. Major carotenoid pigments found were torularhodin, torulene,  $\alpha$ carotene and  $\beta$ -carotene. *X. dendrorhous* was the exception with astaxanthin as the major carotenoid pigment. Maximum carotenoid pigment production (~400  $\mu$ g g<sup>-1</sup>) was achieved with *Rhodotorula mucilaginosa* CRUB 0138 when cultured in fermentor and with the novel species *Cystofilobasidium "lacusmascardii"* CRUB 1046by optimizing culture conditions through a factorial design and response surface analysis. Micosporinogenic strains produced high levels of MGG (up to 48 mg g<sup>-1</sup>) when cultured under PAR+RUV. Extra-cellular enzymatic activities of major importance were aminopeptidase,

lipase and ARNase. The analysis of fatty acids revealed the presence of essential lipids and poli-insaturated fatty acids (PUFAs), comprising mainly palmitic, oleic, linoleic and á-linolenic acids.

The aquatic environments in Patagonia harbor an interesting diversity of pigmented yeasts which includes novel species and strains producing industrially relevant metabolites.

### VII Laboratório de Microbiologia, Departamento de Botânica e Engenharia Biológica, Instituto Superior de Agronomia, 1349-017 Lisboa, Portugal. Communicated by M. Malfeito-Ferreira <mmalfeito@isa.utl.pt>.

Book Chapter.

1 Loureiro, V and Malfeito-Ferreira, M 2006 Spoilage activities of *Dekkera/Brettanomyces* spp.. In: Blackburn, C (ed.) Food spoilage microorganisms. Chapter 13. Woodhead Publishers, Cambridge.

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### VIII Laboratorium voor Microbiologie, Wageningen University, Hesselink van Suchtelenweg 4, 6703 CT Wageningen, The Netherlands. Communicated by W.J. Middelhoven <Wout.Middelhoven@wur.nl>.

1 A paper entitled "Polysaccharides and phenolic compounds as substrate for yeasts isolated from rotten wood and description of *Cryptococcus fagi* Middelhoven et Scorzetti sp. nov. appeared in Antonie van Leeuwenhoek 90:57-67 (2006)." An abstract of this paper already appeared in YNL LIV-II of December 2005.

The following paper has appeared online in Antonie van Leeuwenhoek.

2 WJ Middelhoven and W van Doesburg - Utilization of hexamethylenetetramine (urotropine) by bacteria and yeasts.

A slow growing bacterial population able to utilize hexamethylelenetetramine (urotropine) as sole source of carbon, nitrogen and energy was isolated from soil. Hexamethylenetetramine, systematic name 1,3,5,7-tetraazotricyclo-[3.3.1.1<sup>3,7</sup>]-decane, is a colourless solid compound that is spontaneously formed in an aquous solution of ammonia and formaldehyde. From the crude enrichment culture two bacteria were isolated and identified as *Brevundimonas diminuta* and a *Phyllobacterium* sp. by sequencing of 16S ribosomal DNA. These bacteria also grew

on urotropine but at a lower rate than the enrichment culture. Addition of glucose to the latter resulted in growth of some yeasts that overgrew the bacteria. These were: *Trichosporon laibachii*, *Pichia methanolica* and a yeast type that took an intermediate position between *Leucosporidium scottii* and *Leucosporidiella creatinivora*. Assimilation of urotropine as sole nitrogen source in YCB is very common among yeasts, 46 out of 60 species tested showed this characteristic.

- 3 A manuscript of C.P. Kurtzman and me, describing four novel *Candida* species, two from decaying mushrooms and two from rotten wood, has been submitted for publication in Antonie van Leeuwenhoek.
- 4 Two chapters in books will appear in 2007. They are entitled "Assimilation of unusual carbon compounds" and "Isolation and identification of yeasts", respectively.

### IX Central Control and Testing Institute for Agriculture, Matúśkova 21, 833 15 Bratislava, Slovakia. Communicated by E. Minárik.

Recent publications.

1 Minárik E 2006 Grape must fining after thermovinification of blue grape varieties. Vinařský obzor 99:370 (In Slovak).

Blue grape thermovinification technology may be interesting if it is well managed. It has, however, some unfavorable technological consequences connected with waste fining material. Application of filtration material shows great environmental problems connected by storing and regulations. Alternative solutions after thermovinification are centrifugation, tangential filtration as well as flotation. All technological measures are briefly discussed.

2 Minárik E 2006 Fractional inoculation of grape must by wine yeasts. Vinařský obzor 99:424 (In Slovak).

Recent research demonstrated the advantage of must inoculation by subsequent (fractional) inoculation and not prior to the beginning of alcoholic fermentation by pure yeast starters or autochthous yeasts strains. Starter strains of *Saccharomyces cerevisiae* or *S. uvarum* are usually used. Fractional inoculation is realized when most of the "wild" yeasts are already eliminated.

# X State Scientific-Research Institute for Genetics and Selection of Industrial Microorganisms (GosNIIgenetika), I-Dorozhnyi 1, Moscow 117545, Russia. Communicated by G.I. Naumov and E.S. Naumova <gnaumov@yahoo.com>.

G.I.N. is grateful to the Organising Committee for financial support to participate in the 25th International Specialised Symposium on Yeasts "Systems Biology of Yeasts – from Models to Applications", June 18-21, 2006, Hanasaari, Espoo, Finland. Many thanks to Vladimir Larionov for invitation to give a seminar on yeast diversity at the National Institute of Health/National Cancer Institute (Bethesda, USA, 28 June 2006).

The following are publications for 2006 or in press.

- 1 Naumov GI, Naumova ES, Smith MTh, de Hoog GS 2006 Molecular-genetic diversity of the ascomycetous yeast genus *Arthroascus: Arthroascus babjevae* sp. nov., *Arthroascus fermentans* var. *arxii* var. nov. and geographic populations of *Arthroascus schoenii*. Int J Syst Evol Microbiol 56:1997-2007.
- 2 Naumova ES, Serpova EV, Korshunova IV, Naumov GI 2006 Molecular genetic peculiarities of the yeast *Lachancea kluyveri*. Microbiology (Moscow) (submitted)

Using RFLP analysis and sequencing of non-coding rDNA regions, phylogenetic analysis of  $\beta$ -galactosidase *MEL* genes and molecular karyotyping, we determined a close genetic relatedness of *Lachancea kluyveri* strains isolated in Europe, North America, Japan and Russian Far East. The chromosomal DNAs of *L. kluyveri* ranged in size from 980 to 3100 kb, and haploid number of chromosomes is eight. No correlation was found between the IGS2 restriction profiles, single nucleotide

substitutions in the ITS1/ITS2 rDNA regions and geographical origin of strains and source of their isolation. *L. kluyveri* strains of different origin showed high homology of *MEL* genes: 79–100%. Phylogenetic analysis of all  $\beta$ -galactosidases in the "Saccharomyces" clade suggested certain species specificity of *MEL* genes of the yeasts *L. kluyveri*, *L. cidri*, *Saccharomyces cerevisiae*, *S. paradoxus*, *S. bayanus* and *S. mikatae*.

## 3 Naumov GI 2006 Maltose assimilation in natural strains of *Saccharomyces* cerevisiae depends on respiration. Dokl. Biol. Sci. (submitted).

Natural *S. cerevisiae* strains non-fermenting maltose were found to assimilate this sugar on a special medium. This phenomenon is known as Kluyver effect. It is shown that antimycin A, whis inhibites the respiration, and mitochondrial respiratory-deficient mutations can block maltose assimilation. Comparative analysis of yeast genotypes on MAL genes showed that low-affinity maltose transport is depended on respiration.

- 4 Ivannikova YuV, Serpova EV 2006 Comparative genomics of species Saccharomyces. In: Systems Biology of Yeasts – from Models to Applications, 25th International Specialised Symposium on Yeasts (ISSY25), June 18-21, 2006, Hanasaari, Espoo, Finland, P31.
- 5 Naumov GI, Naumova ES, Martynenko NN 2006 Comparative genomics of *Saccharomyces* yeasts from red berry and grape winemaking. In: Systems Biology of Yeasts – from Models to Applications, 25th International Specialised Symposium on Yeasts (ISSY25), June 18-21, 2006, Hanasaari, Espoo, Finland, P32.
- 6 Naumova ES, Korhola M 2006 Heterozygosity of molecular markers in industrial *Saccharomyces* strains – a new basis for yeast breeding. In: Systems Biology of Yeasts – from Models to Applications, 25th International Specialised Symposium on Yeasts (ISSY25), June 18-21, 2006, Hanasaari, Espoo, Finland, P33.
- 7 ES Naumova defended her dissertation "Evolutionary and taxonomic genetics of yeasts" for the degree of Doctor of Biological Sciences, on June 6, 2006, GosNIIgenetika, Moscow.

YuV Ivannikova has received an INTAS PhD Fellowship (2006-2007) to conduct a collaborative project "Evolutionary genetics of *Saccharomyces* yeasts" with the Faculty of Life Sciences at the University of Manchester, UK (Supervisors: Steve Oliver and Gennadi Naumov).

### XI Faculty of Agronomy, Mendel University of Agriculture and Forestry, Brno, Czech Republic. Communicated by I. Pavelková <Pavelkova.Irena@seznam.cz>.

Recent theses from our laboratory.

1 Bc. Hana Kolínková – Yeast genome analyses methods

The development of molecular-genetic methods is in wide progress nowadays. The detailed study of the yeast genome is extremely important for the recognition of gene sequences as well as whole genetic information. The aim of this remarkable bachelor study was to sum up methods which are being used in yeast biology research, describe them and compare their advantages and disadvantages, taking into consideration their time-consumption and the expenses. The study deals especially with PCR, anaphoresis, sequencing, polymorphism detection and nucleic acids crossing. Furthermore methods such as RFLP, AFLP and RT-PCR are mentioned.

2 Ing. Irena Pavelková - The influence of different active dry yeasts strains on fermentation efficiency of must and quality of wine. Faculty of Horticulture, Lednice na Moravì, Mendel University of Agriculture and Forestry Brno, Czech Republic

Presented thesis provides the view of characteristics of active dry yeasts strains, the advantages of using them in winemaking technology, summarizes the positives and negatives of spontaneous fermentation and compares these methods of winemaking. The thesis emphasizes that the application of active dry yeasts cultures ensures more or less foreseeable fermentation, assures the desirable length of fermentation process and that the residual sugar content shows zero or minimum values. Suggestion that these wines are susceptible to be senzoricaly more identical was not confirmed. It agrees with the supporters of spontaneous fermentation at the point of senzorical diversity and richness of such wines. Nevertheless, on the other hand, to make wines of desirable properties, the combination of many factors – location of vineyard, climate, agronomical practices and last but not least professional treatment of grapes and must - is indispensable.

Key words: yeasts, *Saccharomyces cerevisiae*, fermentation efficiency, wine aroma, quality of wine.

# XII Cell and Organism Biology, Lund University, Solvegatan 35, 22362 Lund, Sweden. Communicated by J. Piškur <Jure.Piskur@cob.lu.se>.

Recent publications.

1 M Mentel, M Špírek, D Jørck-Ramberg, J Piškur In press Transfer of genetic material between pathogenic and foodborne yeasts. Appl Environ Microbiol.

2 Zameitat E, Gojkovic Z, Knecht W, Piskur J, Loffler M 2006 Biochemical characterization of recombinant dihydroorotate dehydrogenase from the opportunistic pathogenic yeast *Candida albicans*. FEBS J. Jun 15; (E-published ahead of print).

*Candida albicans* is the most prevalent yeast pathogen in humans, and recently it has become increasingly resistant to the current antifungal agents. In this study we investigated *C. albicans* dihydroorotate dehydrogenase (DHODH, EC 1.3.99.11), which catalyzes the fourth step of de novo pyrimidine synthesis, as a new target for controlling infection. We propose that the enzyme is a member of the DHODH family 2, which comprises mitochondrially bound enzymes, with quinone as the direct electron acceptor and oxygen as the final electron acceptor. Full-length DHODH and N-terminally truncated DHODH, which lacks the targeting sequence and the transmembrane domain, were subcloned from *C. albicans*, recombinantly expressed in *Escherichia coli*, purified, and characterized for their kinetics and substrate specificity. An inhibitor screening with 28 selected compounds was performed. Only the dianisidine derivative, redoxal, and the biphenyl quinoline-carboxylic acid derivative, brequinar sodium, which are known to be potent inhibitors of mammalian DHODH, markedly reduced *C. albicans* DHODH activity. This study provides a background for the development of antipyrimidines with high efficacy for decreasing in situ pyrimidine nucleotide pools in *C. albicans*.

3 Lohkamp B, Andersen B, Piskur J, Dobritzsch D 2006 The crystal structures of dihydropyrimidinases reaffirm the close relationship between cyclic amidohydrolases and explain their substrate specificity. J Biol Chem 12;281(19):13762-13776.

In eukaryotes, dihydropyrimidinase catalyzes the second step of the reductive pyrimidine degradation, the reversible hydrolytic ring opening of dihydropyrimidines. Here we describe the three-dimensional structures of dihydropyrimidinase from two eukaryotes, the yeast *Saccharomyces kluyveri* and the slime mold *Dictyostelium discoideum*, determined and refined to 2.4 and 2.05 angstroms, respectively. Both enzymes have a (beta/alpha)8-barrel structural core embedding the catalytic di-zinc center, which is accompanied by a smaller beta-sandwich domain. Despite loop-forming insertions in the sequence of the yeast enzyme, the overall structures and architectures of the active sites of the dihydropyrimidinases are strikingly similar to each other, as well as to those of hydantoinases, dihydroorotases, and other members of the amidohydrolase superfamily of enzymes. However, formation of the physiologically relevant

The extension of one of the sheets of the beta-sandwich domain across a subunit-subunit interface in yeast dihydropyrimidinase underlines its closer evolutionary relationship to hydantoinases, whereas the slime mold enzyme shows higher similarity to the noncatalytic collapsin-response mediator proteins involved in neuron development. Catalysis is expected to follow a dihydroorotase-like mechanism but in the opposite direction and with a different substrate. Complexes with dihydrouracil and N-carbamyl-beta-alanine obtained for the yeast dihydropyrimidinase reveal the mode of substrate and product binding and allow conclusions about what determines substrate specificity, stereoselectivity, and the reaction direction among cyclic amidohydrolases.

tetramer shows subtle but nonetheless significant differences.

4 Dobritzsch D, Andersen B, Piskur J. 2005 Crystallization and X-ray diffraction analysis of dihydropyrimidinase from *Saccharomyces kluyveri*. Acta Crystallograph Sect F Struct Biol Cryst Commun. 61:359-362.

Dihydropyrimidinase (EC 3.5.2.2) catalyzes the second step in the reductive pathway of pyrimidine degradation, the hydrolysis of 5,6-dihydrouracil and 5,6-dihydrothymine to the corresponding N-carbamylated beta-amino acids. Crystals of the recombinant enzyme from the yeast *Saccharomyces kluyveri* 

# 5 Piskur J, Rozpedowska E, Polakova S, Merico A, Compagno C. 2006 How did *Saccharomyces* evolve to become a good brewer? Trends Genet 22:183-186.

Brewing and wine production are among the oldest technologies and their products are almost indispensable in our lives. The central biological agents of beer and wine fermentation are yeasts belonging to the genus *Saccharomyces*, which can accumulate ethanol. Recent advances in comparative genomics and bioinformatics have made it possible to elucidate when and diffracting to 2.6 A at a synchrotron-radiation source have been obtained by the hanging-drop vapour-diffusion method. They belong to space group P2(1) (unit-cell parameters a = 91.0, b = 73.0, c = 161.4 A, beta = 91.4 degrees), with one homotetramer per asymmetric unit.

why yeasts produce ethanol in high concentrations, and how this remarkable trait originated and developed during their evolutionary history. Two research groups have shed light on the origin of the genes encoding alcohol dehydrogenase and the process of ethanol accumulation in *Saccharomyces cerevisiae*.

6 Mentel M, Piskur J, Neuveglise C, Rycovska A, Cellengova G, Kolarov J. 2005 Triplicate genes for mitochondrial ADP/ATP carriers in the aerobic yeast *Yarrowia lipolytica* are regulated differentially in the absence of oxygen. Mol Genet Genomics 273:84-91

### 7 Piskur J 2006 Using Fungi as Models. Topics Cur Genet 15 - ISBN: 3-540-31480-6

Fungal comparative genomics started in 2000 by the genome sequencing of several yeast species other than the canonical *Saccharomyces cerevisiae*. Since then, over 30 fungal genome sequences have become available. This set represents a total evolutionary divergence comparable to that between vertebrates and arthropods, but also contains closely related genomes. This volume describes how we can use this set of genomes to trace large and small-scale events in genome evolution, to extract information about highly conserved and less conserved sequence elements, and to develop novel methods in genomics that will have an impact on genomics at large.

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Abstract of a lecture presented at the German Mycological Society meeting at Tuebingen (29th of Sep. till 7th of Oct. 2006).

1 H Prillinger, K Lopandic, T Sugita, M Wuczkowski - *Asterotremella* gen.nov. albida a symbiontic tremelloid yeast isolated from the agarics *Asterophora lycoperdoides* and *A. parasitica*.

Using a genotypic approach (PCR-fingerprinting, DNA/DNA reassociation, partial sequences of the 26S rDNA gene, complete sequences of the 18S rDNA gene, and sequences of the internal transcribed spacer) five tremelloid yeast isolates from the agarics *Asterophora lycoperdoides* and *A. parasitica* were shown to be conspecific with *Cryptococcus* ramirezgomezianus. It was not possible to distinguish the yeast strains from *A. lycoperdoides* and *A. parasitica* using sequences from the intergenic spacer (IGS1). Phylogeny based on the 26S

rDNA (D1/D2-domain) and 18S rDNA demonstrated that *C. ramirezgomezianus* is closely related to several additional *Cryptococcus* species (*C. humicola*, *C. longus*, *C. musci*, *C. pseudolongus*) within the Trichosporonales. A new genus, *Asterotremella*, and a new family, Asterotremellaceae were introduced for *Cryptococcus* species clustering within the Trichosporonales having an ubiquinone Q-9. *Cryptococcus ramirezgomezianus* is a synonym of *Asterotremella albida*.

# XIV 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>.

#### Special Committee on the Nomenclature of Fungi with a Pleomorphic Life Cycle

I was recently invited by Dr. Scott Redhead to join the newly formed "Special Committee on the Nomenclature of Fungi with a Pleomorphic Life Cycle", whose formation was formally announced in Taxon 54: 1057-1064, 2005. This committee reports to the General Committee and the International Botanical Congress in 2011. The Special Committee was set up to report on changes proposed to Article 59 of the International Code of Botanical Nomenclature. The other members of the committee are Drs: Amy Rossman (Chairperson), Scott Redhead (Secretary), Joe Bischoff, Walter Gams, Cheryl Grgurinovic, David Hawksworth, Tsuyoshi Hosoya, Paul Kirk, Keith Seifert, Fred Spiegel and Mike Wingfield.

Article 59 of the International Botanical Code of Nomenclature (ICBN) covers "Names of fungi with a pleomorphic life cycle". At the 17<sup>th</sup> International Botanical Congress held in Vienna last year, the Nomenclatural Section discussed 5 proposals to change Article 59, all initially published by David Hawksworth under the title: Limitation of dual nomenclature for pleomorphic fungi in Taxon 53: 596-598, 2004. These proposals were relabelled 59 proposals A-E in the synopsis published prior to the Congress (Taxon 54: 241-242, 2005). Proposals A and C-E were all withdrawn by the author and simultaneously referred to our Special Committee. Proposal B was emended from the "floor" at the Congress and approved. Proposal B suggested a radical change to Art. 59, to allow for "epitypification" of previously published anamorph names by teleomorphic epitypes, thereby converting the anamorphic name to a holomorphic status, and giving those epitypified names priority status on par with teleomorphic names. The emendations made at the Congress limited the priority from 1 January 2007 onwards, rather than allowing them to be retroactive. The emended wording was published in Taxon 54: 1064, 2005. The new Vienna Code is now published with these changes incorporated.

While the mandate of the Special Committee (Art. 59 committee) is to report on recommendations regarding proposals 59 A, C-E to the XVIII IBC in Melbourne, Australia, in 2011, it is generally recognized by all concerned that this amounts to revisiting the entire premise for and wording of Art. 59.

The following papers have been recently published or are in press.

- 1 Bauer R, Begerow D, Sampaio JP, Weiß M, Oberwinkler F 2006 The simple-septate basidiomycetes: a synopsis. Mycological Progress 5:41-66.
- 2 Gadanho M, Libkind D, Sampaio JP 2006 Yeast diversity in the extreme acidic environments of the Iberian Pyrite Belt. Microbial Ecology (DOI: 10.1007/s00248-006-9027-y).
- 3 Gadanho M, Sampaio JP 2006 Microeukaryotic diversity in the extreme environments of the Iberian Pyrite Belt: a comparison between universal and fungi-specific primer sets, temperature gradient gel electrophoresis and cloning. FEMS Microbiol Ecol 57:139-148.
- 4 Golubev WI, Sampaio JP, Golubeva EW 2006 *Cryptococcus stepposus*, a new filobasidiaceous yeast species found in the Prioksko-terrasny biosphere reserve in Russia. Mycol Res 110:957-961.
- 5 Libkind D, Ruffini A, van Broock M, Alves L, Sampaio JP 2006 Biogeography, host-specificity and molecular phylogeny of the basidiomycetous yeast *Phaffia rhodozyma* and its sexual state *Xanthophyllomyces dendrorhous*. Appl Environ Microbiol (in press).

# XV Department of Microbiology, Institute of Applied Molecular Biology, Saarland University, Postfach 151150, Building A 1.5, D-66041 Saabrücken, Germany. Communicated by M.J. Schmitt <mjs@microbiol.uni-sb.de>.

The following are summaries of papers of the group that have recently been published.

1 Schmitt MJ & Breinig F 2006 Yeast viral killer toxins: lethality and self-protection. Nat Rev Microbiol 4:212-221.

Since the initial discovery of toxin-secreting killer yeasts more than 40 years ago, continuous research on this widespread phenomenon has provided insights into eukaryotic cell biology and virus-host cell interactions. This review will focus on the most recent advances in our understanding of the basic biology of virus-infected killer yeasts, in particular the toxin-encoding killer viruses and the intracellular processing, maturation and toxicity of the viral protein toxins. The strategy of using eukaryotic viral toxins to effectively penetrate and eventually kill a eukaryotic target cell will be discussed, and the cellular mechanisms of self-defence and protective immunity will also be addressed.

2 Breinig F, Sendzik T, Eisfeld K & Schmitt MJ 2006 Dissecting toxin immunity in virus-infected killer yeast uncovers an intrinsic strategy of self-protection. Proc Natl Acad Sci USA 103:3810-3815.

Toxin-secreting 'killer' yeasts were initially identified more than 40 years ago in *Saccharomyces cerevisiae* strains infected with a double-stranded RNA 'killer' virus. Despite extensive research conducted on yeast killer toxins, the mechanism of protecting immunity by which toxin-producing cells evade the lethal activities of these proteins has remained elusive. Here, we identify the mechanism leading to protecting immunity in a killer yeast secreting a viral  $\alpha/\beta$  protein toxin (K28) that enters susceptible cells by receptor-mediated endocytosis and, after retrograde transport into the cytosol, blocks DNA synthesis resulting in both cell cycle arrest and caspase-mediated apoptosis. We demonstrate that toxin immunity is effected within the cytosol of a toxin-secreting yeast and occurs via the formation of complexes between re-internalised toxin and unprocessed precursor moieties (pptox) that are subsequently ubiquitinated and proteasomally degraded, eliminating the active form of the toxin. Interference with cellular ubiquitin homeostasis either through overexpression of mutated ubiquitin (Ub-RR<sup>48/63</sup>) or by blocking deubiquitination, prevents ubiquitination of toxin and results in an impaired immunity and the expression of a suicidal phenotype. The results presented here reveal the uniquely elegant and efficient strategy that killer cells have developed to circumvent the lethal effects of the toxin they produce.

3 Heiligenstein S, Eisfeld K, Sendzik T, Jimenez-Becker N, Breinig F & Schmitt MJ 2006 Retrotranslocation of a viral A/B toxin from the yeast endoplasmic reticulum is independent of ubiquitination and ERAD. EMBO J 25:4717-4727.

K28 is a viral A/B toxin that traverses eukaryotic cells by endocytosis and retrograde transport through the secretory pathway. Here we show that toxin retrotranslocation from the ER requires Kar2p/BiP, Pdi1p, Scj1p, Jem1p and proper maintenance of Ca<sup>2+</sup> homeostasis. Neither cytosolic chaperones nor Cdc48p/Ufd1p/Npl4p complex components or proteasome activity are required for ER exit, indicating that K28 retrotranslocation is mechanistically different from classical ERAD. We demonstrate that K28 exits the ER in a heterodimeric but unfolded conformation and dissociates into its subunits as it emerges into the cytosol where  $\beta$  is ubiquitinated and degraded. ER export and *in vivo* toxicity were not affected in a lysine-free K28 variant nor under conditions when ubiquitination and proteasome activity was blocked. In contrast, toxin uptake from the plasma membrane required Ubc4p (E2) and Rsp5p (E3) and intoxicated *ubc4* and *rsp5* mutants accumulate K28 at the cell surface incapable of toxin internalization. We propose a model in which ubiquitination is involved in the endocytic pathway of the toxin, while ER-to-cytosol retrotranslocation is independent of ubiquitination, ERAD and proteasome activity.

4 Breinig F, Diehl B, Rau S, Zimmer C, Schwab H & Schmitt MJ 2006 Cell surface expression of bacterial esterase A by Saccharomyces cerevisiae and its enhancement by constitutive activation of the cellular unfolded protein response. Appl Environ Microbiol 72:7140-7147.

Yeast cell surface display is a powerful tool in the expression and immobilization of biocatalytically active proteins on a unicellular eukaryote. Here, bacterial carboxylesterase EstA from *Burkholderia gladioli* was covalently anchored into the cell wall of *Saccharomyces cerevisiae* by in frame fusion to the endogenous yeast proteins Kre1p, Cwp2p and Flo1p. By using *p*-nitrophenylacetate as substrate, specific esterase activity of yeast expressing each protein fusion was 103 mU mg<sup>-1</sup> protein for Kre1/EstA/Cwp2p and 72 mU mg<sup>-1</sup> protein for Kre1/EstA/Flo1p. *In vivo* cell wall targeting was confirmed by esterase solubilization after laminarinase treatment and immunofluorescence microscopy. EstA expression resulted in cell wall-associated esterase activity of 2.72 U mg<sup>-1</sup> protein for

Kre1/EstA/Cwp2p and 1.27 U mg<sup>-1</sup> protein for Kre1/EstA/Flo1p. Furthermore, esterase display on the yeast cell surface enabled the cells to effectively grow on the esterase-dependent carbon source glycerol triacetate (Triacetin). In case of Kre1/EstA/Flo1p, *in vivo* maturation within the yeast secretory pathway and final incorporation into the wall was further enhanced under conditions of constitutive activation of the unfolded protein response pathway (UPR). Our results demonstrate that esterase cell surface display in yeast, which as shown here is *per se* remarkably more effective than EstA surface display in *E. coli*, can be further optimized by activating the protein folding machinery within the eukaryotic secretion pathway.

### XVI Department of Food Science and Technology, Process Engineering and Applied Science, Canadian Institute of Fisheries Technology, Dalhousie University, P.O. Box 1000 Halifax, Nova Scotia B3J 2X4, Canada. Communicated by A. Speers. <a href="mailto:<a href="mailto:speers@lal.ca">speers@lal.ca</a>>.

Recent publication.

1 RA Speers, YQ Wan, YL Jin and RJ Stewart 2006 Effects of fermentation parameters and cell wall properties on yeast flocculation. J Inst Brew 112:246–254.

Industrial wort was fermented with a NewFlo phenotype ale yeast in lab-scale cylindrical fermenters. The effects of various fermentation parameters and yeast cell wall properties on yeast flocculation were studied during 120 h fermentation. The evaluation of the cell volume during the fermentation revealed a non-normal distribution (p < 0.05) at most fermentation times. Overall yeast cell size initially decreased during the first 24 h of fermentation then increased during the 24–60 h fermentation period. Cell size subsequently declined until the end of fermentation presumably due to floc settling. While yeast flocculation began after 24 h fermentation, most flocs remained in suspension until 60 h when the average turbulent shear rate caused by CO2 evolution declined to below 8 s–1. Both the Helm's flocculence and cell surface hydrophobicity values rapidly increased to high numbers from 24 h onward. Changes in the orthokinetic capture coefficient ( $\alpha_0$ ) value with fermentation time, measured in fermenting worts, indicated a significant increase (p < 0.001) after 24 h of fermentation. Presumably, this change was due to increases in ethanol and the decline in sugar concentration with time. Although a significant positive correlation (p < 0.05) was observed between zymolectin densities and cell surface areas, the total zymolectin level on yeast cell walls did not change significantly with fermentation time (p > 0.05). Interestingly, no significant difference existed in Helm's flocculation values of suspended and settled yeast cells (p > 0.05). The flocculationrate of LCC125 was readily inhibited by addition of glucose or maltose. Results suggest that fermentable sugar levels and shear force exert major influences on yeast flocculation during beer fermentations.

# XVII Food Microbiology Laboratory, Department of Botany, Sikkim Government College, Gangtok 737102, Sikkim, India. Communicated by Jyoti Prakash Tamang <jyoti\_tamang@hotmail.com>.

We have been working on the microbiology of ethnic fermented foods and beverages of the Himalayan regions of India, Nepal, Bhutan and Tibet in China for last 20 years. The following are out recent publications on that topic.

1 Thapa S and Tamang JP 2004 Product characterization of kodo ko jaanr: fermented finger millet beverage of the Himalayas. Food Microbiol 21:617-622.

*Kodo ko jaanr* is a traditional mild-alcoholic beverage prepared from seeds of finger millets in the Eastern Himalayas. Forty samples of *kodo ko jaanr* were collected from the Darjeeling hills and Sikkim in India and subjected to microbiological and analytical characterisation. Population of yeasts and lactic acid bacteria was detected at the level of 7.1 Log CFU g<sup>-1</sup> and 5.9 Log CFU g<sup>-1</sup>, respectively. Yeasts consisted

of *Pichia anomala*, *Saccharomyces cerevisiae*, *Candida glabrata*, *Saccharomycopsis fibuligera*, and lactic acid bacteria consisted of *Pediococcus pentosaceus* and *Lactobacillus bifermentans* in kodo ko jaanr samples. The pH, moisture, acidity and alcohol content of the product were 4.1, 69.7 %, 0.27 % and 4.8 %, respectively. Kodo ko jaanr is rich in crude fibre.

## 2 Thapa N, Pal J and Tamang JP 2004 Microbial diversity in ngari, hentak and tungtap, fermented fish products of Northeast India. World J Microbiol Biotechnol 20:599-607.

*Ngari, hentak* and *tungtap* are traditional fermented fish products of North East India. Lactic acid bacteria, endospore-forming rods, yeasts and aerobic mesophilic counts ranged from 4.0-7.2, 3.3-4.6, <1-3.5 and 4.3-7.3 log cfu/g, respectively. Yeasts were identified as species of *Candida* and *Saccharomycopsis*. Enzymatic and antimicrobial activities of the

isolates were tested. None of the strains produced biogenic amines in the applied method. Strains of LAB had high degree of hydrophobicity, indicating their 'probiotic' characters. This study has demonstrated the microbial diversity within the species of lactic acid bacteria, *Bacillus* and yeasts.

### 3 Tsuyoshi N, Fudou R, Yamanaka S, Kozaki M, Tamang N, Thapa S and Tamang JP 2005 Identification of yeast strains isolated from marcha in Sikkim, a microbial starter for amylolytic fermentation. Int J Food Microbiol 99:135-146.

*Marcha* is a traditional amylolytic starter used to produce sweet-sour alcoholic drinks, commonly called jaanr in the Himalayan regions of India, Nepal, Bhutan and Tibet (China). The aim of this study was to examine the microflora of *marcha* collected from Sikkim in India, focusing on yeast flora and their roles. Twenty strains of yeasts were isolated from *marcha*, and were identified, based on phenotypic and genotypic characters, as *Saccharomyces bayanus*, *Candida glabrata*, *Pichia anomala*, *Saccharomycopsis fibuligera*, *Saccharomycopsis capsularis* and *Pichia burtonii*. These strains produced ethanol and showed high amylolytic activity.

4 Dewan S and Tamang JP 2006 Microbial and analytical characterization of Chhu, a traditional fermented milk product of the Sikkim Himalayas. J Sci Ind Res 65:747-752.

*Chhu*, an ethnic fermented yak-milk product of Sikkim, contains: lactic acid bacteria (LAB), 8.1-8.8; yeasts, 6.0-6.9; and total viable counts, 8.9-9.2 Log cfu/g. No mould was detected. Yeasts were identified as *Saccharomycopsis crataegensis* and

*Candida castellii*. LAB produced a wide spectrum of enzymes. None of the strains produced bacteriocin and biogenic amines. A proximate composition of *Chhu* was similar to a typical cheese.

# 5 Tamang JP and Thapa S 2006 Fermentation dynamics during production of bhaati jaanr, a traditional fermented rice beverage of the Eastern Himalayas. Food Biotechnol 20:251-261.

Bhaati jaanr is an inexpensive high calorie mild-alcoholic beverage prepared from the steamed glutinous rice, consumed as a staple food beverage in the Eastern Himalayan regions of Nepal, India and Bhutan. In this paper, fermentation dynamics including growth kinetics and physico-chemical changes during fermentation of *bhaati jaanr* were studied. Population of filamentous moulds declined significantly (P<0.05) every day and finally disappeared after the 5<sup>th</sup> day. Load of yeasts increased significantly (P<0.05) from 10<sup>5</sup> cfu/g to 10<sup>8</sup> cfu g<sup>-1</sup> within 2<sup>nd</sup> day. Maximum activities of saccharification and liquefaction of rice were observed on 3<sup>rd</sup> day of fermentation. It was revealed that *Saccharomycopsis fibuligera* and *Rhizopus* spp. play the important roles in saccharification process of rice in *bhaati jaanr* fermentation. The means pH, acidity, moisture and alcohol content of the product were 3.5, 0.24 %, 83.4 % and 5.9 %, respectively.

### XVIII VTT Technical Research Centre of Finland, P.O.Box 1000, FI-02044 VTT, Finland. Communicated by John Londesborough <john.londesborough@vtt.fi>.

Publications since our last communication include the following.

- 1 Guimarães PMR, Virtanen H and Londesborough J 2006 Direct evidence that maltose transport activity is affected by the lipid composition of brewer's yeast. J. Inst. Brew. 112:203-209.
- 2 Ilmén M, Koivuranta K, Ruohonen L, Suominen P, Penttilä M Efficient production of L-lactic acid from xylose by *Pichia stipitis*. Appl. Environ. Microbiol. (in press).

- 3 Rintala E, Pitkänen JP, Vehkomäki ML, Penttilä M, Ruohonen L 2006 The ORF *YNL274c* (*GOR1*) codes for glyoxylate reductase in *Saccharomyces cerevisiae*. Yeast (*in press*).
- 4 Saloheimo A, Rauta J, Stasyk OV, Sibirny AA, Penttilä M, Ruohonen, L Xylose transport studies with xylose-utilizing *Saccharomyces cerevisiae* strains expressing heterologous and homologous permeases. Appl. Microbiol. Biotechnol. (in press).

### XIX Department of Biology, University of Western Ontario, London, Ontario, Canada N6A 5B7. Communicated by M.A. Lachance <a>(a)uwo.ca>.

The following paper listed in the Spring issue of the YNL is now in print. It was featured in the November issue of Microbiology Today in the "Hot off the press" section.

1 Lachance MA, Bowles JM, Wiens F, Dobson J, and Ewing CP 2006 *Metschnikowia orientalis* sp. nov., an Australasian yeast from nitidulid beetles. Int J Syst Evol Microbiol 56:2489-2493.

The following have been accepted recently.

2 Rosa CA, Lachance MA, Teixeira LCRS, Pimenta RS & Morais PB 2007 *Metschnikowia cerradonensis* sp. nov., a yeast species isolated from ephemeral flowers and their nitidulid beetles in Brazil. Int J Syst Evol Microbiol (in press).

A novel yeast species, *Metschnikowia cerradonensis* sp. nov., is described from 12 strains isolated from flowers of *Ipomoea carnea* and from beetles of the genus *Conotelus* in the Cerrado ecosystem in the region of Jalapao, Tocantins State, Brazil. Analysis of the sequences of the rRNA gene cluster suggested that *M. cerradonensis* is closely related to *Metschnikowia santaceciliae*, *Metschnikowia continentalis* and an undescribed species represented by strain UWOPS 00-154.1. These species mate together but ascospores are very rarely formed, showing that they represent distinct biological species. *M. cerradonensis* is apparently endemic to the Cerrado ecosystem of the Jalapao area. The type strain of M. cerradonensis is UFMG 03-T67.1<sup>T</sup> (h<sup>+</sup>) (= CBS 10409<sup>T</sup>=NRRL Y-48067T) and the designated allotype is UFMG 03-T68.1 (h") (=CBS 10410=NRRL Y-48068).

3 Manson JS, Lachance MA, Thomson JD In press *Candida gelsemii* sp. nov., a yeast of the Metschnikowiaceae clade isolated from nectar of the poisonous Carolina jessamine. Antonie van Leeuwenhoek (in press).

A new yeast species, *Candida gelsemii*, 11 is described to accommodate three isolates recovered in Georgia, USA, from the toxic nectar of the Carolina jessamine (*Gelsemium sempervirens*). The species resembles other members of the Metschnikowiaceae clade that have been recovered from nectar, but differs in a number of morphological and physiological

characteristics. Analysis of rDNA sequences places the new species well into the clade, but in a basal position with respect to a group of *Metschnikowia* and *Candida* species known to occur in association with nectars, bees, as well as marine invertebrates. The type is strain UWOPS 06–24.1T (CBS 10509<sup>T</sup>, NRRL - 48212<sup>T</sup>).

4 Lachance MA In press Current Status of *Kluyveromyces* Systematics. FEMS Yeast Res (special issue on *Kluyveromyces*).

A brief outline of the current taxonomic status of the genus *Kluyveromyces* is presented. Noteworthy are the transfer of several former *Kluyveromyces* species to other genera, the

conservation of the name *Kluyveromyces* for *K. lactis*, *K. marxianus*, and four related species, and some recent attempts to clarify the variety status of strains assigned to *K. lactis*.

I shall present the following poster in January, in the 'ecogeographic rules' session, at the 3<sup>rd</sup> Conference of the International Biogeography Society, in Tenerife, Canary Islands.

5 Lachance MA 2007 Yeast biogeography and the ubiquity debate.

One currently advocated interpretation of Beijerinck's principle, "everything is everywhere", is that the microbial world consists of a relatively small number of very widely distributed species and consequently that the study of microbial biogeography is a vain pursuit (e.g., Fenchel & Finlay 2004 Bioscience 54:777-784). Here, I explore the question of ubiquity

in two groups of related yeasts that were sampled extensively across the globe. Recent advances in sequence-based yeast identification methods have caused an explosion in the number of yeast species descriptions and have allowed biogeographic hypotheses to be tested. Yeasts assigned to the plant decay-associated *Sporopachydermia* clade belong to 21 phylotypes, 17 of which probably represent separate species. A single species, *S. lactativora* might be construed as cosmopolitan; in all other cases, the phylotypes have distributions that can be attributed to vicariance or dispersal, and many appear to be endemic. Likewise, a large collection of *Metschnikowia* and related species isolated from ephemeral flowers and their nitidulid beetles or other insects can be assigned clearly to 22 biological species whose distributions follow patterns that are consistent with biogeographic history. Endemism predominates,

although some cosmopolitan species also exit. Of special interest is the recent discovery that endemic beetles that live on endemic plants of Hawai'i harbour at least six endemic sister species of *Metschnikowia*. In addition, yeast species found in ephemeral flowers and associated insects exhibit a latitudinal species richness gradient. Taken together, these results provide strong support to the view that in effect, many microorganisms are subject to the rules of historical biogeography.

## **Obituaries** Carl F. Robinow - 1909-2006

Carl Franz Robinow, 97, a faculty member for 26 years with the Department of Microbiology & Immunology [University of Western Ontario], passed away October 20. He had retired from Western in July, 1975. The service for Robinow was private. Robinow enjoyed a productive career as Professor of Microbiology, a researcher of note in bacterial and fungal cytology, and as a stimulating and humorous companion to a wide range of colleagues, friends and students. He was born in Hamburg, Germany on April 10, 1909. Schooling in Germany was followed by medical studies in clinical centres in Freiburg and Vienna, attaining his M.D. in Hamburg in 1934. Robinow pursued research in microbiology, first in Copenhagen, Denmark, moving later to London, England where he met and married Rosie Derenburg. Following WW II, they lived for two years in the United States. He came to Canada in 1949 via an appointment in Western's then-department of bacteriology and immunology in the faculty of medicine. The department he joined was small and busy with an active clinical laboratory, a teaching program for medical students, and a growing research program with graduate students. He did more than his share in all aspects of academic life in a lively post-war medical school. He was awarded the Harrison Prize in 1957 and elected a Fellow of the Royal Society of Canada in 1960. He was president of the

Canadian Society of Microbiologists in 1962 and of the Canadian Society for Cell Biology in 1968; he was appointed professor emeritus in 1978, and awarded an honorary D.Sc. by Western in 1983. The death of his wife in 1972 and his retirement in 1974 seemed not to alter his research productivity for he carried on his work for more than 25 years thereafter. He would never avoid a cytological challenge as happened when he was consulted by E. A. Angert about how to study the structure of a remarkable very large bacterium that reproduces by forming multiple new cells in its cytoplasm. They published a paper in 1998 when he was 89, another venture in comparative cytology. No less remarkable for a retiree was the collaborative paper "The bacterial nucleoid revisited" with E. Kellenberger in 1994 and the major review (noted above) in 2002. His health deteriorated following a fall in 2003 and medical and surgical adventures led to life in a nursing home. Friends speak fondly of Robinow's nature which has been described as a blend of the scientific and the romantic, enlivened by a puckish sense of humour. He was proud of the fact that his long life permitted him to view Halley's Comet twice, first in 1911 and on its return in 1986. He is mourned by his sons, John Anthony Robinow and Richard Oliver Robinow; daughter-in-law Gwen Thompson; and grandchildren Richard and Elizabeth.

### Bob Klanac

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### Some Personal Notes on Dr Robinow

I had the privilege of crossing paths with Dr Robinow many times. The first encounter was in 1973, when he was a guest speaker at the Université de Montréal. I was then an MSc student at Macdonald College. A graduate student colleague had suggested that we travel downtown to see and hear this famous researcher who, among other things, had succeeded in photographing the bacterial chromosome and demonstrating the presence of three chromosomes in *Schizosaccharomyces pombe*. As he did every time I had the occasion to attend one of his seminars, Dr Robinow delighted his audience with a combination of 35 mm and large glass lantern slides, and of course the two projectors required to show them. Six years later, I met him personally at the time of my job interview at the University of Western Ontario. He greeted me warmly and took me to his laboratory to show me fascinating organisms in his various microscopes. This was the first of many sessions, some of which lasted from sunrise to sunset, spent discussing yeasts, nuclei, chromosomes, the wavelength of light, all this interspersed with stories of his visits to the Davis and Paris labs where I had studied. In 1980, Dr Robinow passed on to me the responsibility of teaching the mycology section of our introductory microbiology course. Although I continued teaching the lectures and laboratory exercises for many years, I never felt the need to make any major changes to the topics covered or the materials presented in the lab exercises, which were outstanding.

One of the very nice outcomes of Dr Robinow's longevity was the frequency at which we celebrated his retirement. The first such event for me was the Carl Robinow Symposium on Microbial Structure and Function, held in 1980. The most recent was a celebration of his 90<sup>th</sup> birthday, during which he gave a research seminar on the strange mitosis of a protist

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endowed with a hundred chromosomes.

I also met Dr Robinow at concerts, where he never failed to tell me a relevant story. For example, after a piano recital, he informed me that his grandmother had been a student of Clara Schumann. He appreciated music. Upon visiting him in the hospital after his fall, I found him (and the rest of the ward) being charmed by the stunning music of daughter-in-law, the distinguished violinist Gwen Thompson.

Dr Robinow had his favourite eating places in London (Ontario), and Jane and I were fortunate enough to join him on several occasions for dinner at the Cafédu-Midi or the Marienbad, where he delighted us with stories about great microbiologists, microscopes, and his World War II tribulations. But the highlight was his response to a waiter offering him the cork for his evaluation, which he executed expertly by holding the cork to his ear and declaring: "this wine sounds excellent!".

Dr Robinow was a founding member of the International Commission on Yeasts and an esteemed colleague to many readers of the Yeast Newsletter.

### Václav Švejcar - 1924-2006

Czech oenologist Ing. Václav Švejcar, CSc. was a skilled horticulturist who later on gained his knowledge of Vitis vinifera L. and winemaking at the Pomological Institute in Ruzynì. After graduating from the University of Agriculture in Prague, he taught in secondary agricultural schools in Milník (Czech Republic) and Modra (Slovakia). In 1961 he was appointed lecturer at the University of Agriculture in Lednice na Moravì, where he stayed until his retirement 1989. Švejcar also served as inspector of agricultural schools. In 1967 he presented his thesis, Study and Classification of Yeast Flora in Vineyards of University of Agriculture in Lednice na Moravì. He conducted many of research projects dealing with yeasts, their

### Irena Pavelková

ecology, biology, and fundamental importance in winemaking. He was enthusiastic advocate of the use of active dry yeasts cultures. He published over 30 scientific publications, some in foreign countries, as well as many scientific and popular articles, reviews and reports. He co-authored books on viticulture and winemaking. In addition, wrote many university papers, often as a co-author with Doc. Ing. Erich Minárik, DrCs. Švejcar was a member of the Microbiological Society of the Czechoslovak Academy of Science and a member of Board for the Valuation of Grape-Vines and the Czech Association of Consumer Cooperatives. He served on the editorial board of two horticultural and viticultural journals.

### International Commission on Yeasts Meeting of Commissioners, June 20th, 2006, ISSY 25 – Hanasaari - Espoo, Finland Minutes of Meeting

**Present:** Leda C. Mendonça-Hagler (Chair), Graham Fleet (IUMS-Mycology Chair), Merja Penttilä, Matti Korhola, José M. Peinado, Patrizia Romano, Doris Rauhut, A. Sibirny, Monique Bolotin-Fukuhara, Gennadi Naumov, Hana Sychrova.

**Apologies**: L. Scheffers (Vice-Chair), I. Spencer-Martins, P. Biely, S. Meyer, A. Martini, A. Vaughan-Martini.

**Report from the Chair**: Leda C. Mendonça-Hagler welcomed the delegates to the meeting and gave apologies for those who could not attend. She expressed her appreciation to Prof. Merja Penttilä and her group for the excellent organization of ISSY 25 and their support for ICY meeting. She presented the agenda and entertained requests for the inclusion of any additional items.

**New Commissioners**: Prof. Valentyn Pidhorsky (Ukraine), Dr. Ivan Hapala (Slovakia) and Prof. Lisa Granchi (Italy) were nominated with a warm welcome from the commissioners.

**Minutes of the previous meeting:** L. Mendonça-Hagler reported on the ICY meeting which took place during ISSY 24, on October 1<sup>st</sup>, 2005, at Hotel Marina d'Or, Oropesa del Mar, Spain. The minutes for this last meeting were published in the December 2005 issue of the Yeast Newsletter and it was sent by electronic mail to the commissioners.

### **Reports on Meetings:**

ISSY 24 (2005) - Oropesa del Mar, Spain, Sep 28 - Oct 2, Genomics, Proteomics and Functional Analysis: A tribute to the scientific career of José Ruiz-Herrera. Organized by Prof. R. Sentandreu. Prof. J. Peinado reported on ISSY 24, which was attended by some 180 delegates. On behalf of ICY, L. Mendonça-Hagler expressed her gratitude to the Spanish group for the successful meeting, mentioning the program's high level and the Spanish warm hospitality.

ISSY 25 (2006) - Hanasaari, Espoo, Finland, June 18 - 21, Systems Biology of Yeasts - from Models to Applications. The meeting was organized by Prof. M. Penttilä. She reported on the ongoing ISSY 25, which was attended by some 240 delegates and had the participation of 40 speakers. Prof. M. Penttilä commented on the high attendance, which was limited by the facilities at Hanasaari. She also stressed the excellent scientific quality of the abstracts. The attendance of substantial proportion of young investigators was noted with appreciation. On behalf of ICY, L. Mendonça-Hagler expressed her gratitude to the organizers for their dedication and commented on the good choice of the seaside location. She proposed a toast to Prof. Prof. M. Penttilä and the organizers, for the excellent work done to realize this highly appreciated symposium. A special issue of FEMS Yeast Research will include contributions presented at ISSY 25.

ISSY 26 (2007) - June 3 - 7, Sorrento, (Naples), Italy: From alcoholic beverages to bioethanol - a new challenge for fermenting yeast. Website: Prof. Patrizia Romano is http://www.issy26.org. organizing this Symposium. She reported on the preparations for the meeting and announced the venue: Hotel Vesuvio, located in Sorrento and 48 km from Naples. This Symposium aims to stress the central role of the yeast Saccharomyces cerevisiae in the production of bioethanol for transportation and update actual knowledge on its use as starter in the alcoholic beverages industry. The relevant contribution of Prof A. Martini, an active member of the organizing committee, was stressed. During the closing ceremony of ISSY 25, Prof. Romano did a presentation announcing ISSY 26 and inviting the audience to visit Italy.

ICY 2008 - 12th International Congress on Yeasts, Kiev, Ukraine. During ICY meeting in Budapest (2003) Prof. A. Sibirny proposed to have the International Yeast Congress (ICY 2008), in Kiev. A progress report was presented by A. Sibirny during ICY 2004, in Rio de Janeiro. At this ICY meeting, Prof. Sibirny reported on the negotiations to get the best choice for a location. He mentioned the ongoing contacts with sponsors and discussed the topics to be addressed. He also announced a presentation on Kiev highlights, which was delivered during the ISSY 25 closing ceremony.

**ISSY 27 (2009) - France**. Prof. M. Bolotin-Fukuhara proposed to organize ISSY 27 in France. The proposal was welcomed by the Commissioners during ICY 2004 in Rio de Janeiro. During the meeting in Espoo, Prof. Bolotin-Fukuhara mentioned the decision to dedicate the symposium to the memory of Louis Pasteur. The well deserved tribute received unanimous acclamation by ICY commissioners. She also reported the necessity to look for a venue some years in advance. Therefore, the French group is already examining the economic feasibility of several locations to hold ISSY 27. **ISSY 28 (2010) - Thailand.** Proposed by Dr. C. Charoenchai. L. Mendonça-Hagler reported on a letter of intent received from Dr. C. Charoenchai regarding the organization of a symposium in Thailand. The proposal was accepted by the Commissioners at ICY meeting in Oropesa del Mar, Spain. No further report was presented at this meeting.

**Report on IUMS meetings:** Prof Graham Fleet, Chair of the Mycology Division of IUMS, reported on the Congress IUMS 2005, which included the 11<sup>th</sup> International Congress of Mycology, held in San Francisco, July 24-29<sup>th</sup>. The congress was hosted by the American Society for Microbiology. Prof. Fleet devoted considerable effort to achieve a good balance between topics on yeasts and filamentous fungi. IUMS 2005 was well attended by ICY commissioners. For the occasion, L. Mendonça-Hagler reported on current ICY activities. ICY was recognized by IUMS as one of the more active COMCOFs. Prof. Fleet reported on the ongoing preparations for IUMS 2008 to be held in Istambul, Turkey (4<sup>th</sup>-15<sup>th</sup> of August), wishing for an increased attendance and successful organization. M. Penttilä proposed a toast to him in recognition to the efforts on behalf of yeast scientific community.

**Report on YNL:** Leda Mendonça-Hagler mentioned the relevant contribution of Prof. André Lachance in managing the **Yeast Newsletter**. She took the opportunity to encourage the commissioners to send reports of their research groups to YNL and to visit the home page:

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

**Other business and activities**: No further topics were included in the agenda.

Adjournment: On behalf of ICY, Leda Mendonça-Hagler closed the meeting expressing her gratitude to the organizers for the successful ISSY 25, held by seaside at Hanasaari Center, Espoo. She stressed her appreciation to M. Penttilä, M. Korhola, and the local team for arranging the epicurean lunch. All present expressed their satisfaction with ISSY 25 with remarks regarding the friendly atmosphere in Finland and the luck of having a week of sunny weather.

Leda C. Mendonça-Hagler

## Recent meetings

### 34<sup>th</sup> Annual Conference on Yeasts of the Czech and Slovak Commission for Yeasts Smolenice, Slovakia, May 10-12, 2006

The 34th Annual Conference on Yeasts took place in the Smolenice Castle, the Congress center of the Slovak Academy of Sciences. The Czech and Slovak Commission for Yeasts and the Institute of Chemistry, Slovak Academy of Sciences, housing the large yeast collection, were again the main organizers of this traditional meeting of the Czech and Slovak yeast researchers. The opening ceremony of the conference was connected with presentation of the František Patočka Medal to Dr. Peter Biely, former chair of the Yeast Commission, for his scientific achievements and his long-year activities in the Commission. The Medal is the highest award of the Czechoslovak Society for Microbiology and was presented to Dr. Biely by members of the Society Committee, chaired by Prof. Ing. Ivan Čižnár, DrSc. This year the conference had the character of an international meeting. Besides Czech and Slovak participants numbering 68, there were a dozen participants from Hungary, Poland, Austria and France. Consequently, the conference language was English. The scientific program consisted of lectures in three sessions, which were complemented by 49 posters.

The titles of lectures and posters are listed below.

## Lectures in the Yeast Cytology and Biochemistry session

- 1 Hašek J Yeast Fluorescence Microscopy. (Invited Lecture in the memory of Dr. Kocková-Kratochvílová)
- 2 Wolinski H, Heise B, Klement EP, Kohlwein SD (Austria). Live cell imaging at the limits of optical resolution.
- 3 Zlámalová M, Hendrych T, Pichová A, Gášková D, Sigler K Identifying interactions of bioactive compounds with S, cerevisiae MDR pumps.
- 4 Czabany T, Athenstaedt K, Wagner A, Müllner H, Sorger D, Daum G (Austria). Dynamics of neutral lipid synthesis and mobilisation in yeast.
- 5 Mazáň M, Farkaš V Transglutaminase activity is involved in the formation of *Saccharomyces cerevisiae* cell wall.

### Lectures in the Biotechnology session

- 6 Thévenieau F, Nicaud, JM, Gaillardin C (France). Hydrophobic substrate utilization by the nonconventional yeast *Yarrowia lipolitica* and its potential.
- 7 Kočí R, Kubešová J, Hulínová T, Márová I Exogenous stress influence on biotechnological production of pigments by red yeasts.
- 8 Rapta P, Zalibera M, Čertík M, Breierová E Carotenogenic yeasts stressed by Cu2+ ions - an exceptional case in EPR studies of antioxidant capacity of yeast extracts.
- 9 Pajtinka M, Sejáková Z, Kogan G, Dercová K Sorption capacities of fungal polysaccharides of cell walls for PCP.
- 10 Klišová T, Patáková P, Heřman J. Image analysis of yeasts.
- 11 Brezová V, Čertík M, Brindzová L, Rapta P, Staško A Analysis of phenolic antioxidants in red wines and EPR studies of their radical scavenging activity.
- 12 Krátky J, Bobček R Effect of esterified glucomannan on milk quality.
- 13 Fuchsberger M Significant trifles.

### Special lecture with wine tasting:

14 Vajcziková I Sensorial evaluation of wines from the Rača region (Slovakia).

## Lectures in the Yeasts genetics and molecular biology session

- 15 Sipiczki M, Miklos I, Szilagyi Z, Batta G (Hungary) General transcription regulators (MEDIATOR and SAGA) and fork-head transcription factors in the regulation of cytokinesis in *Schizosaccharomyces pombe*.
- 16 Antunovics Z, Sipiczki M (Hungary) Genome stabilization and rearrangement in interspecific hybrids Saccharomyces cerevisiae X Saccharomyces uvarum.
- 17 Holešová Z, Kosa P, Ghindea R, Tomáška Ľ, Nosek J. Screening for Genes Implicated in The Control of Morphogenesis of the Pathogenic Yeast *Candida parapsilosis*.
- 18 Bialková A, Šubík J Biology of the yeast *Candida* glabrata.
- 19 Autengruber A, Zlámalová M, Pichová A, Sigler K Aging in yeast as reflected in changes in cells' respiration pattern.

- 20 Kinclová-Zimmermannová O, Zavřel M, Sychrová H. Amino-acid residues involved in determination of substrate specificity of yeast plasma membrane Na+/H+ antiporters.
- 21 Janitor M Site-directed mutagenesis in yeast research.

### Posters

- 22 Puchart V, Vršanská M, Biely P Yeast enzymes for preparation of chromogenic substrates for endoxylanases.
- 23 Kogan G, Miadoková E, Rauko P, Svidová S, Vlčková V, Dúhová V, Naïová S Glucomannan of *Candida utilis* – a versatile biomodulatory polysaccharide.
- 24 Mrózová Z, Czabany T, Špaņovį M, Valachovič M, Čertík M, Hapala I. "Slim yeast on high-fat diet": Saccharomyces cerevisiae as a model in obesity research?
- 25 Maceková D, Farkaš V Ecto-glycanase activities in *Cryptococcus neoformans*.
- 26 Šimočková M, Holič R, Griač P Putative phosphatidylglycerol specific phospholipase C in yeast *Saccharomyces cerevisae*.
- 27 Tahotná D, Holič R, Poloncová K, Šimočková M, Griač P Phospholipid transfer activity of yeast Sec14p is not essential for its function in vivo.
- 28 Tomšíková A The present problems of candidoses.
- 29 Vlčková V, Kopásková M, Dúhová V, Hlavová M, Špinerová L, Naďová S, Miadoková E The antigenotoxic activity of the extract from artichoke *Cynara cardunculus* L. in yeast and other model systems.
- 30 Čertík M, Brindzová L, Rapta P, Brezová V, Vajcziková I Antioxidant and radical scavenging ability od red wines from Raca region (Slovakia) with high content of natural polyphenols.
- 31 Omelková J, Matalová S, Klučáková M, Sláviková E Utilization of lignite and soil humic acids by yeasts and yeast – like fungi.
- 32 Sláviková E, Vadkertiová R Yeasts colonizing the leaf surfaces.
- 33 Breierová E, Gregor T, Marová I, Čertík M, Kogan G Enhanced antioxidant formula based on a selenium supplemented carotenoid producing yeast biomass.

- 34 Raclavský V, Moráňová Z, Husičková V, Novotný R, Pavlíček J, Kawamoto S Further characterization of *Cryptococcus neoformans* hypoxia response.
- 35 Cieśla M, Balicki K, Towpik J, Boguta M Mafldirected repression of RNA polymerse III is dependent on carbon source.
- 36 Džugasová V, Baťová M, Šubík J Phenotypic analysis of *Candida glabrata* mutant strains displaying altered fluconazole susceptibility.
- 37 Čierna M, Fekete V, Míčková A, Sulo P Revised occurrence of Petite positive species in *Saccharomyces clade*.
- 38 Fekete V, Hapala I, Sulo P Dependance of mitstate on the presence of Different ADP/ATP translocator forms. (Construction of nearly petite negative *Saccharomyces cerevisiae* strain).
- 39 Bialková A. Breunig K, Gbelská Y Inventory of genes involved in multidrug resistance in five related yeast species.
- 40 Gunišová, S, Kramara, J, Nosek, J, Tomáška Ľ Are there two forms of TaZ1 in vivo? How are they regulated?
- 41 Bartoníčková L, Ptáčková P, Kraidlová L, Palková Z, Janderová B. The effect of Ccr4-Not complex on S. cerevisiae colonies.
- 42 Kinský S, Gunišová S, Tomáška Ľ DNA binding protein Bas1 and its functions in *Yarrowia lipolytica*.
- 43 Kiššová I, Deffieu M, Salin B, Velours G, Manon S, Camougrand N Mitochondria - selective autophagy (mitophagy) in yeast.
- 44 Gavenčiaková B, Kosa P, Nosek J A set of plasmid vectors for genetic manipulations of the pathogenic yeast *Candida parapsilosis*.
- 45 Laco J, Zeman I, Kolarov J Investigation of the physiological and molecular functions of the mitochondrial carrier Sal1p.
- 46 Machula K, Sosinska G, Palamarczyk G Tryptophan to glycine substitution in dolichol binding site of the yeast dolichol kinase affects the cell wall.
- 47 Habovštiaková J, Tichá E, Obernauerová M Functional state of mitochondria and *Saccharomyces cerevisiae* cell susceptibility to external stress factors.
- 48 Papousková K, Sychrová H Yeast plasma membrane alkali metal cation/H+ antiporters: A phylogenetic study.

- 49 Višacká, K, Petrezsélyová, S, Gunišová, S, Nosek, J, Tomáška Ľ. Camthmg: Mitochondrial protein of *Candida albicans* with potential role in MTDNA stabilization.
- 50 Keszthelyi A, Farkas Z, Hamari Z, Kucsera J, Pfeiffer I Killer toxins against pathogenic yeasts.
- 51 Pichová A, Regl G, Breitenbach M, Sigler K DNA damage in replicative aging of *Saccharomyces cerevisiae*.
- 52 Drobcová B, Mentel M, Kiššová I, Kolarov J, Polčic P. BH3-only Proteins in the Bcl-2 family as studied in yeast.
- 53 Patrášová M, Poliaková- Košťanová D, Čertík M, Šabová Ľ The role of cardiolipin and phospatidylglycerol in Bax mediated cytotoxicity in *K.lactis.*
- 54 Poliaková-Košť anová D, Dunajčíková P, Patrášová M, Šabová Ľ A role for cytochrome C in cytotoxic effect of Bax in *K.lactis*: creation a mutant strain resistant to Bax.
- 55 Ptáčková P, Bartoníčková L, Janderová B, Palková Z Development of *S. cerevisiae* colonies monitored by the expression of ACT1, SSA3 and CCR4 genes.
- 56 Stromájer-Rácz T, Gazdag Z, Benkõ Z, Antal J, Pesti M. Oxidative stress induced by a mutant viral protein R (VPR) of HIV-1 expressed in *Schizosaccharomyces pombe*.
- 57 Tóth D.M, Gazdag Z, Pesti M Characterization of a tert-butyl hydroperoxide tolerant mutant of *Schizosaccharomyces pombe*.
- 58 Šarinová M, Straková V, Gbelská Y Functional state of mitochondria and its influence on membrane properties in *Kluyveromyces lactis*.
- 59 Sidorová M, Hikkel I, Drobná E, Šubík J Galactose induced expression of pdr3- alleles decreases the accumulation of rhodamine 6G and enhances the susceptibility of gain-of-function pdr1 mutants to antifungals.
- 60 Laurenčík M, Jakubová R, Vajcziková I. Breierová E, Čertík M, Franko F, Sulo P Yeasts "troublemakers" in Vinea and wine.
- 61 Marešová L, Sychrová H Complementation of *S. cerevisiae* kha1 deletion phenotypes by a plant antiporter AtCHX17.
- 62 Vránová D, Vadkertiova R The application of PCR-RFLP as a Taxonomic Tool in yeasts identification in food.

- 63 Kaminska J, Sedek M, Wysocka-Kapcinska M, Zoladek T A yeast actin cytoskeleton-associated protein, Pan1, could be a shuttle protein.
- 64 Pribylova L, de Montigny J, Sychrova H. Genetic manipulation in the osmotolerant yeast *Zygosaccharomyces rouxi*.
- 65 Kinclová-Zimmermannová O, Sychrová H The Cnh1 antiporter is important for potassium and pH homeostasis in *C. albicans* cells.
- 66 Pevala V, Kolarov J The cell death in fission yeast, prevented with Bcl-xL protein, is accompanied with changes of mitochondrial morphology and physiology.
- 67 Drábková M, Márová I, Kubešová J, Halienová A, Čarnecká M, Kočí R Isolation and characterization of some subcellular fractions of stressed red yeasts.
- 68 Márová I, Obruča S, Ondruška V, David J, Vojtová L, Babák L, Jančář J Biodegradation of modified polyuretane foams by *Aureobasidium pullulans* and by termophillic bacteria: a pilot comparative study.

- 69 Kubešová J, Turková V, Mikulcová A, Kučerík J, Márová I, Pekař M Use of *Saccharomyces cerevisiae* D7 to analysis of genotoxicity/ antimutagenicity of processed humic acids.
- 70 Sigler K, Kosař K, Mikyška A, Gabriel P, Dienstbier M Assessing and predicting yeast vitality by the acidification power test: critical reassessment.

At the meeting of the Czech and Slovak Commission for Yeasts held at the end of the conference it was decided that the 35<sup>th</sup> Annual conference on Yeasts will be held again in the Smolenice Castle during May 16-18, 2007 and its main themes will be Genetics and Molecular Biology, Cell Biology and Biochemistry, Biotechnology, and Medical Mycology. The conference language will be English and will be open to foreign scientist. Yeast researchers particularly from the neighboring countries are expected in increasing numbers. The information on the future conferences can be found on the web site <u>www.chem.sk/yeast</u>.

Communicated by Peter Biely

## **Forthcoming Meetings**

## 35<sup>th</sup> Annual Conference on Yeasts - Smolenice, Slovakia, May 16<sup>th</sup> - 18<sup>th</sup> 2007

On-line registration is still open at http://www.chem.sk/yeast in December 2006. Main topics:

- 1 Genetics and molecular biology of yeasts
- 2 Cell biology and biochemistry of yeasts
- 3 Biotechnology and medical mycology

The deadline for Registration and Accommodation reservation is March 15<sup>th</sup> 2007. For Abstract submission the deadline is March 31<sup>st</sup> 2007.

### Yeast 2007 Melbourne Exhibition and Convention Centre - July 1-6, 2007 <u>www.yeast2007.org</u>

Yet another of the world's best genetics conferences is coming to Australia! This time it will be the 23<sup>rd</sup> International Conference on Yeast Genetics and Molecular Biology (ICYGMB). I would argue that yeast is the best model for genetics research and it is certainly a well known contributor to molecular biology. Even the latest Nobel Prize in Chemistry goes to Roger Kornberg is for discoveries made in yeast.

This meeting is held every second year and it will be the first time it has been held in Australia. It is expected to attract around 1000 yeast researchers from around the globe and is eagerly anticipated by our local researchers, known as The Australian Yeast Group

[www.australianyeastgroup.org]. The program for the meeting is well advanced and will include Keynote speakers Gerry Fink and Sir Paul Nurse as well as 30 symposia speakers.

Conference themes include:

Yeasts in brewing, wine and biotechnology Protein transport and turnover Membrane proteins and lipids Other yeast and fungi as model systems Cytoskeleton

Yeasts as pathogens: biology and clinical concerns Post-translational modifications and proteomics Transcription and control of gene expression Chromosomes - structure and inheritance Organelle division and inheritance Cell signalling Yeast models for human disease and ageing Bioinformatics and genome-wide studies

Nuclear structure/ organization

There will be considerable public engagement with the final day devoted to sessions on the contributions of yeast to our lives in the 21st century. Topics will include:

Communicated by Wieland Meyer

The supply of insulin produced in yeast to diabetics Prevention of liver cancer and hepatitis by yeast-derived vaccines New vaccine for cervical cancer The role of yeast in cancer research Yeast in neurodegenerative disease research Yeast in the screening of new drugs Yeast in the development of a malaria vaccine New therapeutic antibodies for cancers from yeast Yeast and contributions to the energy crisis

Please keep informed up updates on our website and register your interest to attend. We look forward to seeing you there.

Ian Macreadie (Conference Chair, on behalf on The Australian Yeast Group)