Yeast

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http://www.uwo.ca/biology/YeastNewsletter/Index.html

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

M.A. Lachance

I School of Life Sciences, Institute of Life Sciences and Green Development, Hebei University, Baoding, 071002, Hebei, China. Communicated by Qi-Ming Wang <wangqm@hbu.edu.cn>.

Recent publications.

1 Jiang YL, Bao WJ, Liu F, Wang GS, Yurkov A.M, Ma Q, Hu ZD, Chen XH, Zhao WN, Li AH, Wang QM. 2024. Proposal of one new family, seven new genera and seventy new basidiomycetous yeast species mostly isolated from Tibet and Yunnan provinces, China. Stud Mycol 109:57-153 - https://doi.org/10.3114/sim.2024.109.02

2 Zhao YJ, Ma Q, Liu MM, Wang QM. 2024. *Malassezia gallinae* sp. nov., a new basidiomycetous yeast species isolated from skins of chickens. Med Mycol 62(11):myae109 - https://doi.org/10.1093/mmy/myae109

3 Liu M-M, Zhu H-H, Bai J, Tian Z-Y, Zhao Y-J, Boekhout T, Wang Q-M. 2024. Breast cancer colonization by *Malassezia globosa* accelerates tumor growth. mBio 15(10):e0199324 - https://doi.org/10.1128/mbio.01993-24

II VTTTechnical Research Centre of Finland, Tietotie 2, P.O. Box 1000, 02044 VTT, Espoo, Finland. Communicated by Kristoffer Krogerus <Kristoffer.Krogerus@vtt.fi>.

Recent publication.

1 Preiss R., Fletcher E. Garshol LM, Garsholz LM, Fosters B, Ozsahin E, Lubberts M, van der Merwe G, Krogerus K. 2024. European farmhouse brewing yeasts form a distinct genetic group. Appl Genet Microbiol Biotechnol 108:430 - https://doi.org/10.1007/s00253-024-13267-3

III Laboratory of Genetics, Wisconsin Energy Institute, DOE Great Lakes Bioenergy Research Center, Center for Genomic Science Innovation, J. F. Crow Institute for the Study of Evolution, University of Wisconsin, Madison, WI 53726, USA. Communicated by Chris Todd Hittinger <cthittinger@wisc.edu>.

Madison, WI, USA, was chosen by the International Commission on Yeasts to host the International Congress on Yeasts in 2028! We hope to see you there. Details will follow.

Recent publications.

1 Crandall JG, Zhou X, Rokas A, Hittinger CT. 2004. Specialization restricts the evolutionary paths available to yeast sugar transporters. Mol Biol Evol 41(11):msae228- https://doi.org/10.1093/molbev/msae228

Functional innovation at the protein level is a key source of evolutionary novelties. The constraints on functional innovations are likely to be highly specific in different proteins, which are shaped by their unique histories and the extent of global epistasis that arises from their structures and biochemistries. These contextual nuances in the sequence-function relationship have implications both for a basic understanding of the evolutionary process and for engineering proteins with desirable properties. Here, we have investigated the molecular basis of novel function in a model member of an ancient, conserved, and biotechnologically relevant protein family. These Major Facilitator Superfamily sugar porters are a functionally diverse group of proteins that are thought to be highly plastic and evolvable. By dissecting a recent evolutionary innovation in an α-glucoside transporter from the yeast *Saccharomyces eubayanus*, we show that the ability to transport a novel substrate requires high-order interactions between many protein regions and numerous specific residues proximal to the transport channel. To reconcile the functional diversity of this family with the constrained evolution of this model protein, we generated new, state-of-the-art genome annotations for 332 Saccharomycotina yeast species spanning approximately 400 million years of evolution. By integrating phylogenetic and phenotypic analyses across these species, we show that the model yeast α-glucoside transporters likely evolved from a multifunctional ancestor and became subfunctionalized. The accumulation of additive and epistatic substitutions likely entrenched this subfunction, which made the simultaneous acquisition of multiple interacting substitutions the only reasonably accessible path to novelty.

2 Zavala B, Dineen L, Fisher KJ, Opulente DA, Harrison MC, Wolters JF, Shen XX, Zhou X, Groenewald M, Hittinger CT, Rokas A, LaBella AL. 2024. Genomic factors shaping codon usage across the Saccharomycotina subphylum. G3 14:jkae207 - https://doi.org/10.1093/g3journal/jkae207

Codon usage bias, or the unequal use of synonymous codons, is observed across genes, genomes, and between species. It has been implicated in many cellular functions, such as translation dynamics and transcript stability, but can also be shaped by neutral forces. We characterized codon usage across 1,154 strains from 1,051 species from the fungal subphylum Saccharomycotina to gain insight into the biases, molecular mechanisms, evolution, and genomic features contributing to codon usage patterns. We found a general preference for A/T-ending codons and correlations between codon usage bias, GC content, and tRNA-ome size. Codon usage bias is distinct between the 12 orders to such a degree that yeasts can be classified with an accuracy >90% using a machine learning algorithm. We also characterized the degree to which codon usage bias is impacted by translational selection. We found it was influenced by a combination of features, including the number of coding sequences, BUSCO count, and genome length. Our analysis also revealed an extreme bias in codon usage in the Saccharomycodales associated with a lack of predicted arginine tRNAs that decode CGN codons, leaving only the AGN codons to encode arginine. Analysis of Saccharomycodales gene expression, tRNA sequences, and codon evolution suggests that avoidance of the CGN codons is associated with a decline in arginine tRNA function. Consistent with previous findings, codon usage bias within the Saccharomycotina is shaped by genomic features and GC bias. However, we find cases of extreme codon usage preference and avoidance along yeast lineages, suggesting additional forces may be shaping the evolution of specific codons.

3 Liu C, Zhou X, Li Y, Hittinger CT, Pan R, Huang J, Chen XX, Rokas A, Chen Y, Shen XX. 2024. The influence of the number of tree searches on maximum likelihood inference in phylogenomics. Syst Biol 73:807-22 - https://doi.org/10.1093/sysbio/syae031

Maximum likelihood (ML) phylogenetic inference is widely used in phylogenomics. As heuristic searches most likely find suboptimal trees, it is recommended to conduct multiple (e.g., 10) tree searches in phylogenetic analyses. However, beyond its positive role, how and to what extent multiple tree searches aid ML phylogenetic inference remains poorly explored. Here, we found that a random starting tree was not as effective as the BioNJ and parsimony starting trees in inferring the ML gene tree and that RAxML-NG and PhyML were less sensitive to different starting trees than IQ-TREE. We then examined the effect of the number of tree searches on ML tree inference with IQ-TREE and RAxML-NG, by running 100 tree searches on 19,414 gene alignments from 15 animal, plant, and fungal phylogenomic datasets. We found that the number of tree searches substantially impacted the recovery of the best-of-100 ML gene tree topology among 100 searches for a given ML program. In addition, all of the concatenation-based trees were topologically identical if the number of tree searches was ≥10. Quartet-based ASTRAL trees inferred from 1 to 80 tree searches differed topologically from those inferred from 100 tree searches for 6/15 phylogenomic datasets. Finally, our simulations showed that gene alignments with lower difficulty scores had a higher chance of finding the best-of-100 gene tree topology and were more likely to yield the correct trees.

4 Rojas J, Hose J, Auguste Dutcher H, Place M, Wolters JF, Hittinger CT, Gasch AP. 2024. Comparative modeling reveals the molecular determinants of aneuploidy fitness cost in a wild yeast model. Cell Genom 4:100656 - https://doi.org/10.1016/j.xgen.2024.100656

Although implicated as deleterious in many organisms, aneuploidy can underlie rapid phenotypic evolution. However, aneuploidy will be maintained only if the benefit outweighs the cost, which remains incompletely understood. To quantify this cost and the molecular determinants behind it, we generated a panel of chromosome duplications in Saccharomyces cerevisiae and applied comparative modeling and molecular validation to understand aneuploidy toxicity. We show that 74%-94% of the variance in aneuploid strains' growth rates is explained by the cumulative cost of genes on each chromosome, measured for single-gene duplications using a genomic library, along with the deleterious contribution of small nucleolar RNAs (snoRNAs) and beneficial effects of tRNAs. Machine learning to identify properties of detrimental gene duplicates provided no support for the balance hypothesis of aneuploidy toxicity and instead identified gene length as the best predictor of toxicity. Our results present a generalized framework for the cost of aneuploidy with implications for disease biology and evolution.

5 Harrison MC, Opulente DA, Wolters JF, Shen XX, Zhou X, Groenewald M, Hittinger CT, Rokas A, LaBella AL. 2024. Exploring Saccharomycotina yeast ecology through an ecological ontology framework. Yeast 41:615-28 - https://doi.org/10.1002/yea.3981

Yeasts in the subphylum Saccharomycotina are found across the globe in disparate ecosystems. A major aim of yeast research is to understand the diversity and evolution of ecological traits, such as carbon metabolic breadth, insect association, and cactophily. This includes studying aspects of ecological traits like genetic architecture or association with other phenotypic traits. Genomic resources in the Saccharomycotina have grown rapidly. Ecological data, however, are still limited for many species, especially those only known from species descriptions where usually only a limited number of strains are studied. Moreover, ecological information is recorded in natural language format limiting high throughput computational analysis. To address these limitations, we developed an ontological framework for the analysis of yeast ecology. A total of 1,088 yeast strains were added to the Ontology of Yeast Environments (OYE) and analyzed in a machine-learning framework to connect genotype to ecology. This framework is flexible and can be extended to additional isolates, species, or environmental sequencing data. Widespread adoption of OYE would greatly aid the study of macroecology in the Saccharomycotina subphylum.

6 Gonçalves C, Harrison MC, Steenwyk JL, Opulente DA, LaBella AL, Wolters JF, Zhou X, Shen XX, Groenewald M, Hittinger CT, Rokas A. 2024. Diverse signatures of convergent evolution in cacti-associated yeasts. PLoS Biol 22:e3002832 - https://doi.org/10.1371/journal.pbio.3002832

Many distantly related organisms have convergently evolved traits and lifestyles that enable them to live in similar ecological environments. However, the extent of phenotypic convergence evolving through the same or distinct genetic trajectories remains an open question. Here, we leverage a comprehensive dataset of genomic and phenotypic data from 1,049 yeast species in the subphylum Saccharomycotina (Kingdom Fungi, Phylum Ascomycota) to explore signatures of convergent evolution in cactophilic yeasts, ecological specialists associated with cacti. We inferred that the ecological association of yeasts with cacti arose independently approximately 17 times. Using a machine learning-based approach, we further found that cactophily can be predicted with 76% accuracy from both functional genomic and phenotypic data. The most informative feature for predicting cactophily was thermotolerance, which we found to be likely associated with altered evolutionary rates of genes impacting the cell envelope in several cactophilic lineages. We also identified horizontal gene transfer and duplication events of plant cell wall-degrading enzymes in distantly related cactophilic clades, suggesting that putatively adaptive traits evolved independently through disparate molecular mechanisms. Notably, we found that multiple cactophilic species and their close relatives have been reported as emerging human opportunistic pathogens, suggesting that the cactophilic lifestyle-and perhaps more generally lifestyles favoring thermotolerance-might preadapt yeasts to cause human disease. This work underscores the potential of a multifaceted approach involving high-throughput genomic and phenotypic data to shed light onto ecological adaptation and highlights how convergent evolution to wild environments could facilitate the transition to human pathogenicity.

7 de Lima AEP, Coplien J, Anthony LC, Sato TK, Zhang Y, Karlen SD, Hittinger CT, Maravelias CT. 2024. On the synthesis of biorefineries for high-yield isobutanol production: from biomass-to-alcohol experiments to system level analysis. RSC Sustain 2:2532-40 - https://doi.org/10.1039/D4SU00283K

The production of isobutanol from lignocellulose has gained attention due to its favorable physical and chemical properties. The use of lignocellulosic biomass as a feedstock to produce isobutanol has substantial sustainability benefits, but the biological conversion to isobutanol faces challenges, such as low yields and by-product formation. In this work, we demonstrate the high-yield production of isobutanol through microbial fermentation of pulp hydrolysates. Three hydrolysates are produced from poplar, sorghum, and switchgrass using pretreatment based on γ-valerolactone. Furthermore, we synthesize a biomass-to-isobutanol biorefinery and perform technoeconomic analysis of three resulting processes using experimental results obtained from an engineered yeast strain which consumes most of the glucose available in the hydrolysate and produces isobutanol at 89–94% theoretical yields. The corresponding minimum fuel selling price (MFSP) is $14.40–$16.01 per gasoline gallon equivalent, with the sorghum-based biorefinery resulting in the lowest price. We identify that solvent/biomass ratio during pretreatment and enzyme loading during hydrolysis have the greatest impact on the MFSP; improvements in these parameters can reduce the MFSP by 46%.

8 Aranguiz K, Horianopoulos LC, Elkin L, Abá KS, Wrobel RL, Shiu SH, Rokas A, Hittinger CT. Machine learning reveals genes impacting oxidative stress resistance across yeasts. bioRxiv - https://doi.org/10.1101/2024.08.14.607963

Reactive oxygen species (ROS) are highly reactive molecules encountered by yeasts during routine metabolism and during interactions with other organisms, including host infection. Here, we characterized the variation in resistance to ROS across the ancient yeast subphylum Saccharomycotina and used machine learning (ML) to identify gene families whose sizes were predictive of ROS resistance. The most predictive features were enriched in gene families related to cell wall organization and included two reductase gene families. We estimated the quantitative contributions of features to each species' classification to guide experimental validation and showed that overexpression of the old yellow enzyme (OYE) reductase increased ROS resistance in *Kluyveromyces lactis*, while *Saccharomyces cerevisiae* mutants lacking multiple mannosyltransferase-encoding genes were hypersensitive to ROS. Altogether, this work provides a framework for how ML can uncover genetic mechanisms underlying trait variation across diverse species and inform trait manipulation for clinical and biotechnological applications.

9 David KT, Schraiber JG, Crandall JG, Labella AL, Opulente DO, Harrison MC, Wolters JF, Zhou X, Shen XX, Groenewald M, Hittinger CT, Pennell M, Rokas A. Convergent expansions of keystone gene families drive metabolic innovation in a major eukaryotic clade. bioRxiv - https://doi.org/10.1101/2024.07.22.604484

Many remarkable innovations have repeatedly occurred across vast evolutionary distances. When convergent traits emerge on the tree of life, they are sometimes driven by the same underlying gene families, while other times many different gene families are involved. Conversely, a gene family may be repeatedly recruited for a single trait or many different traits. To understand the general rules governing convergence at both genomic and phenotypic levels, we systematically tested associations between 56 binary metabolic traits and gene count in 14,710 gene families from 993 species of *Saccharomycotina* yeasts. Using a recently developed phylogenetic approach that reduces spurious correlations, we discovered that gene family expansion and contraction was significantly linked to trait gain and loss in 45/56 (80%) of traits. While 601/746 (81%) of significant gene families were associated with only one trait, we also identified several 'keystone' gene families that were significantly associated with up to 13/56 (23%) of all traits. These results indicate that metabolic innovations in yeasts are governed by a narrow set of major genetic elements and mechanisms.

10 Feng B, Li Y, Liu H, Steenwyk JL, David KT, Tian X, Xu B, Gonçalves C, Opulente DA, LaBella AL, Harrison MC, Wolters JF, Shao S, Chen Z, Fisher KJ, Groenewald M, Hittinger CT, Shen XX, Rokas A, Zhou X, Li Y. Unique trajectory of gene family evolution from genomic analysis of nearly all known species in an ancient yeast lineage. bioRxiv - https://doi.org/10.1101/2024.06.05.597512

Gene gains and losses are a major driver of genome evolution; their precise characterization can provide insights into the origin and diversification of major lineages. Here, we examined gene family evolution of 1,154 genomes from nearly all known species in the medically and technologically important yeast subphylum Saccharomycotina. We found that yeast gene family and genome evolution are distinct from plants, animals, and filamentous ascomycetes and are characterized by small genome sizes and smaller gene numbers but larger gene family sizes. Faster-evolving lineages (FELs) in yeasts experienced significantly higher rates of gene losses—commensurate with a narrowing of metabolic niche breadth—but higher speciation rates than their slower-evolving sister lineages (SELs). Gene families most often lost are those involved in mRNA splicing, carbohydrate metabolism, and cell division and are likely associated with intron loss, metabolic breadth, and non-canonical cell cycle processes. Our results highlight the significant role of gene family contractions in the evolution of yeast metabolism, genome function, and speciation, and suggest that gene family evolutionary trajectories have differed markedly across major eukaryotic lineages.

IV State Scientific-Research Institute for Genetics and Selection of Industrial Microorganisms (GosNIIgenetika), NRC "Kurchatov Institute", I-Dorozhnyi 1, Moscow 117545, Russia. Communicated by E.S. Naumova <lena_naumova@yahoo.com>.

The following are papers for 2024 or in press:

1 Tuaeva AYu, Ponomareva AM, Livshits VA, Naumova ES. 2024. Yeast microflora of dairy products sold in Russia. Microbiology (Moscow) 93(5):629–639.

2 Tuaeva AYu, Kachmazov GS, Naumova ES. 2024. Peculiarities of the yeast microflora of authentic Ossetia cheeses. Biotechnologiya 40(1):109–114 (in Russian).

Yeast microflora of brined Ossetia cheeses made from raw milk by small farms in various regions of North and South Ossetia has been studied, for the first time. A total of 17 yeast species belonging to 10 genera were identified. It is shown that the composition of the yeast microflora of Ossetia cheeses depends on the place of production, the microflora of raw milk and starter culture, as well as on the type of milk. Most of the studied Ossetian cheeses were dominated by lactose-utilizing yeasts *Kluyveromyces lactis* and *Debaryomyces hansenii*, as well as by lactose-negative yeasts *Saccharomyces cerevisiae*, *Pichia kudriavzevii* and *P. fermentans*. The composition of the microflora of artisanal Ossetia cheeses is more diverse than that of commercial cheeses produced from pasteurized milk.

3 Borovkova AN, Shalamitskiy MYu, Naumova ES. 2024. Comparative genetic analysis of pectinase genes *PGU* of the yeast *Saccharomyces cerevisiae*: selection of strains with high pectinolytic activity. Biotechnologiya, 40 (5) (in press) (in Russian).

A large-scale screening of pectinolytic activity was carried out in *Saccharomyces cerevisiae* strains isolated from different types of wines, grapes, fermenting must, fruit and berry juices, as well as from natural sources and industrial waste in different regions of the world. Of 305 strains studied, the majority (249) did not produce active endo-polygalacturonase. The results indicate that the ability to hydrolyze pectin is generally not characteristic of *S. cerevisiae* yeasts. Regardless of the source and place of isolation, *S. cerevisiae* strains have a single pectinase gene *PGU1*, as a rule, located on the chromosome X. Another chromosomal localization of *PGU1* genes was found only in four strains isolated from fermented palm juice in Malaysia and wine strain I-411 (Transcarpathia, Ukraine): chromosomes VII/XV and IX. Ten strains of *S. cerevisiae* with the highest pectinolytic activity were selected, which are of interest for further molecular genetic research and breeding work with wine yeasts.

V Ecology Department, Lawrence Berkeley National Laboratory, One Cyclotron Road, Berkeley, CA 94720. Communicated by Tamas Torok <ttorok@lbl.gov>.

Recent publications.

1 Bijlani et al. 2022. Genomic characterization of the Titan-like cell producing *Naganishia tulchinskyi*, the first novel eukaryote isolated from the international space station. J Fungi (MDPI) 8(2):165 - https://doi.org/10.3390/jof8020165

Multiple strains of a novel yeast belonging to genus *Naganishia* were isolated from environmental surfaces aboard the International Space Station (ISS). Confocal, scanning, and transmission electron microscopy revealed distinct morphological differences between the microgravity-grown cells and the standard Earth gravity-grown cells. Phylogenetic analyses indicated that these ISS strains represented a single species in the genus *Naganishia* and were clustered with *Naganishia diffluens*. The name *Naganishia tulchinskyi* is proposed to accommodate these strains, with IF6SW-B1^T as the holotype.

2 Leo et al. 2023. Genomic characterization and radiation tolerance of *Naganishia kalamii* sp. nov. and *Cystobasidium onofrii* sp. nov. from Mars 2020 mission assembly facilities. IMA Fungus 14:15 - https://doi.org/10.1186/s43008-023-00119-4

During the construction and assembly of the Mars 2020 mission components at two different NASA cleanrooms, several fungal strains with yeast-like appearance were isolated. The polyphasic taxonomic approach supported the conclusion that the two basidiomycetous yeasts belong to hitherto undescribed species. We propose two novel species with the type strains, *Naganishia kalamii* sp. nov. (FJI-L2-BK-P3^T = NRRL 64466 = DSM 115730) and *Cystobasidium onofrii* sp. nov. (FKI-L6-BK-PAB1^T = NRRL 64426 = DSM 114625). The genomic analysis predicted proteins associated with dehydration and desiccation stress-response and the presence of genes that are directly related to osmotolerance and psychrotolerance. Both novel species were also UV resistant.

3 Leo et al. 2023. Draft genome sequencing of *Naganishia* species isolated from the polar environments. Microbiol Resour Announc 12(10):e0038823 - https://doi.org/10.1128/MRA.00388-23

The draft genomes of five *Naganishia* strains were sequenced using MinION and annotated using Funannotate pipeline. Phylogenetic and genomic analyses were performed to provide their genetic relationships, diversity, and potential functional capabilities. This approach will aid in understanding their potential to survive under microgravity and their resilience to extreme environments.

VI Lodz University of Technology. Faculty of Biotechnology and Food Sciences, Department of Environmental Biotechnology, Wolczanska 171/173, 90-530 Lodz, Poland. Communicated by Prof. Dorota Kregiel <dorota.kregiel@p.lodz.pl>.

The following papers have been published.

1 Sobolewska E, Borowski S, Kręgiel D. 2024. Cultivation of yeasts on liquid digestate to remove organic pollutants and nutrients and for potential application as co-culture with microalgae. J Environmental Management 362:121351 - https://doi.org/10.1016/j.jenvman.2024.121351

In this study, the growth of yeast and yeast-like fungi in the liquid digestate from vegetable wastes was investigated in order to remove nutrients and organic pollutants, and for their application as co-culture members with green microalgae. The studied yeast strains were characterized for their assimilative and enzymatic profiles as well as temperature requirements. In the first experimental stage, the growth dynamics of each strain were determined, allowing to select the best yeasts for further studies. In the subsequent stage, the ability of selectants to remove organic pollutants was assessed. Different cultivation media containing respectively 1:3, 1:1, 3:1 vol ratio of liquid digestate and the basal minimal medium were used. Among all tested yeast strains, *Rhodotorula mucilaginosa* DSM 70825 showed the most promising results, demonstrating the highest potential for removing organic substrates and nutrients. Depending on the medium, this strain achieved 50–80% sCOD, 45–60% tVFAs, 21–45% TN, 33–52% PO_4^{3-} reduction rates. Similar results were obtained for the strain *Candida* sp. OR687571. The high nutrient and organics removal efficiency by these yeasts could likely be linked to their ability to assimilate xylose (being the main source of carbon in the liquid digestate). In culture media containing liquid digestate, both yeast strains achieved good viability and proliferation potential. In the liquid digestate medium, *R. mucilaginosa* and *Candida* sp. showed vitality at the level of 51.5% and 45.0%, respectively. These strains seem to be a good starting material for developing effective digestate treatment strategies involving monocultures and/or consortia with other yeasts or green microalgae.

2 Kregiel D, Krajewska A, Kowalska-Baron A, Czarnecka-Chrebelska KH, Nowak A. 2024. Photoprotective effects of yeast pulcherrimin. Molecules (MDPI) 29(20):4873 - https://doi.org/10.3390/molecules29204873

Sunscreen products can protect the skin against the harmful effects of UV radiation, including reddening, aging, and cancer. The aim of this research was to evaluate the photoprotective effects of yeast pulcherrimin, an iron-chelating dipeptide. We first investigated the cytotoxicity of pulcherrimin produced by *Metschnikowia pulcherrima* yeast on the human keratinocyte HaCaT cell line, using the PrestoBlue assay. We assessed the ability of pulcherrimin to induce DNA repair after the exposure of HaCaT cells to oxidative stress. We also evaluated its protective activity against UVC radiation. The sun protective factor (SPF) was calculated using the Mansur equation. The UVA/UVB ratio values for pure pulcherrimin were evaluated using the Boots Star Rating system. The critical wavelength was determined by calculating the integrated optical density curve area. Based on the results, pulcherrimin shows strong cytoprotective effects through antioxidant and photoprotective activities on the HaCaT cell line. The calculated SPFs were 20 and 15 at pH = 7 and pH = 10, respectively. The critical wavelength above 370 nm and the UVA/UVB ratio R > 1 suggest that yeast pulcherrimin - a cyclic dipeptide containing iron - may be considered a promising photoprotective agent.

VII Department of Chemistry and Biotechnology, School of Science, Tallinn University of Technology, Tallinn, Estonia and Lallemand Inc, Montréal, Canada. Communicated by Ildar Nisamedtinov <inisamedtinov@lallemand.com>.

Recent publication.

1 Berga HY, Arjuc G, Nisamedtinov I. 2024. Nitrogen availability and utilisation of oligopeptides by yeast in industrial scotch grain whisky fermentation. J Amer Soc Brew Chem - https://doi.org/10.1080/03610470.2024.2389608

Scotch grain whisky is produced with a substantial proportion of unmalted grains, which can result in nitrogen deficiency for yeast in the fermentable grain mash. This study examined nitrogen source availability and utilisation by three commercial whisky strains (*Saccharomyces cerevisiae*) during Scotch grain whisky fermentation, focusing on oligopeptides. Peptide uptake kinetics in synthetic whisky mash with defined peptides showed that oligopeptides of up to nine amino acids were taken up by the strains, albeit with some variability between the strains. The study found that peptides with appropriate molecular weights could replace free amino acids without negatively affecting fermentation kinetics. Moreover, fermentation performance improved when additional nitrogen was provided via peptides rather than diammonium phosphate. Analysis of industrial grain mash indicated that despite low initial yeast assimilable nitrogen, residual proteolytic activity from malt increased nitrogen availability during fermentation. Approximately 30% of the nitrogen consumed by yeast during grain mash fermentation was derived from peptides. LC-HRMS peptide analysis revealed complex dynamics of peptide formation, degradation, and utilisation. This study highlights the importance of oligopeptides in ensuring optimal fermentation efficiency in Scotch grain mash and similar substrates.

VIII Department of Soil Biology, Faculty of Soil Science, Lomonosov Moscow State University, 119234, Leninskie gory, 1/12, Moscow, Russia, and All-Russian Collection of Microorganisms (VKM), G.K. Skryabin Institute of Biochemistry and Physiology of Microorganisms RAS, 142290, pr. Nauki 5, Pushchino, Russia. Communicatedd by A.V. Kachalkin <kachalkin_a@mail.ru>.

Recent publications

1 Glushakova A, Tepeeva A, Prokof'eva T, Kachalkin A. 2024. Culturable yeast diversity in urban topsoil influenced by various anthropogenic impacts. International Microbiology 27:1383–1403 - https://doi.org/10.1007/s10123-024-00482-1

In urban ecosystems, processes associated with anthropogenic influences almost always lead to changes in soil micromycete complexes. The taxonomic structure of soil micromycete complexes is an important informative parameter of soil bioindication in the ecological control of urban environments. Unicellular fungi, such as culturable yeasts, are a very suitable and promising object of microbiological research for monitoring urban topsoil. This review aims to give an overview of the yeast communities in urban topsoil in different areas of Moscow (heating main area, household waste storage and disposal area, highway area) and to discuss the changes in the taxonomic structure of culturable yeast complexes depending on the type and intensity of anthropogenic impact.

2 Sonets IV, Solovyev MA, Ivanova VA, Vasiluev PA, Kachalkin AV, Ochkalova SD, Korobeynikov AI, Razin SV, Ulianov SV, Tyakht AV. 2024. Hi-C metagenomics facilitate comparative genome analysis of bacteria and yeast from spontaneous beer and cider. Food Microbiology 121:104520 - https://doi.org/10.1016/j.fm.2024.104520

Sequence-based analysis of fermented foods and beverages' microbiomes offers insights into their impact on taste and consumer health. High-throughput metagenomics provide detailed taxonomic and functional community profiling, but bacterial and yeast genome reconstruction and mobile genetic elements tracking are to be improved. We established a pipeline for exploring fermented foods microbiomes using metagenomics coupled with chromosome conformation capture (Hi-C metagenomics). The approach was applied to analyze a collection of spontaneously fermented beers and ciders (n = 12). The Hi-C reads were used to reconstruct the metagenome-assembled genomes (MAGs) of bacteria and yeasts.

3 Kachalkin A, Tomashevskaya M, Pankratov T, Yurkov A. 2024. Endothallic yeasts in the terricolous lichens *Cladonia*. Mycological Progress 23:29 - https://doi.org/10.1007/s11557-024-01966-0

Lichens are multi-component microbial communities where yeasts are common. This work was designed to address the taxonomic diversity and distribution of cultivated endothallic yeasts in terricolous lichens of the genus *Cladonia.* Lichens *Cladonia stellaris*, *C. rangiferina*, *C. cornuta*, and *C. pleurota* from arctic, subarctic, and continental boreal climates in Russia in a range of biotopes were studied. In total, 40 yeast species were isolated. It revealed a broad taxonomic range of endothallic yeast with a substantial proportion of basidiomycetes from subphyla *Agaricomycotina* and *Pucciniomycotina*. Many common yeast species were found within the epithallic and endothallic yeast communities of some lichens studied. The physiological and ecological characterization of 30 basidiomycetous endothallic yeasts enhances our understanding of lichen habitats and may aid in culturing seldom-isolated lichenicolous fungi. Out of 40 species in this study, 21 new yeast species were discovered, and 10 new yeast species were proposed with their formal descriptions. They are *Colacogloea glushakovae*, *Cyrenella lichenicola*, *Microsporomyces wangii*, *Microsporomyces cladoniae*, *Genolevuria nadymea*, *Teunia turchettiae*, *Phaeotremella sibirica*, *Phaeotremella endothallina*, *Piskurozyma altaica*, and *Piskurozyma cladoniicola*.

4 Glushakova A, Sharova A, Kachalkin A. 2024. Seasonal dynamics of culturable yeasts in ornithogenically influenced soils in a temperate forest and evaluation of extracellular enzyme secretion in *Tausonia pullulans* at different temperatures. Journal of Fungi (MDPI) 10(8):532 - https://doi.org/10.3390/jof10080532

The culturable yeast communities in temperate forest soils under the ornithogenic influence were studied in a seasonal dynamic. It was found that the yeast abundance in the soil under strong ornithogenic influence reached the highest values in winter compared to the other seasons and amounted to 4.8 lg (cfu/g). This was almost an order of magnitude higher than the minimum value of yeast abundance in ornithogenic soils determined for summer. A total of 44 yeast species, 21 ascomycetes and 23 basidiomycetes, were detected in ornithogenic soil samples during the year. These included soil-related species (*Barnettozyma californica*, *Cyberlindnera misumaiensis*, *Cutaneotrichosporon moniliiforme*, *Goffeauzyma gastrica*, *Holtermanniella festucosa*, *Leucosporidium creatinivorum*, *L. yakuticum*, *Naganishia adeliensis*, *N. albidosimilis*, *N. globosa*, *Tausonia pullulans*, and *Vanrija albida*), eurybionts (yeast-like fungus *Aureobasidium pullulans*, *Debaryomyces hansenii*, and *Rhodotorula mucilaginosa*), inhabitants of plant substrates and litter (*Cystofilobasidium capitatum*, *Cys. infirmominiatum*, *Cys. macerans*, *Filobasidium magnum*, *Hanseniaspora* *uvarum*, *Metschnikowia pulcherrima*, and *Rh. babjevae*) as well as a group of pathogenic and opportunistic yeast species (*Arxiozyma bovina*, *Candida albicans*, *C. parapsilosis*, *C. tropicalis*, *Clavispora lusitaniae*, and *Nakaseomyces glabratus*). Under an ornithogenic influence, the diversity of soil yeasts was higher compared to the control. Interestingly, the absolute dominant species in ornithogenic soils in winter (when the topsoil temperature was below zero) was the basidiomycetous psychrotolerant yeast *T. pullulans*. Screening of the hydrolytic activity of 50 strains of this species at different temperatures (2, 4, 10, 15 and 20 °C) showed that the activity of esterases, lipases and proteases was significantly higher at the low cultivation temperature.

5 Kachalkin A, Bekkarevich A, Tomashevskaya M, Glushakova A. 2024. Yeasts from frass of longhorn beetle larvae (Cerambycidae) in birch wood and description of *Diddensiella monakovoensis* f.a, sp. nov. Biologia 79:3219–3226 - https://doi.org/10.1007/s11756-024-01770-x

In the study of yeasts from frass of longhorn beetle larvae, species with high abundance from the genera *Diddensiella, Scheffersomyces* and *Sugiyamaella* and some others with lower abundance were observed. The novel ascomycetous yeast species *Diddensiella monakovoensis* f.a, sp. nov. (holotype: KBP Y-6906; isotypes: VKM Y-3676 and DBVPG 8067) is proposed based on a conventional DNA-barcode sequence analysis and physiological characteristics.

6 Glushakova A, Kachalkin A. 2024. Diversity of culturable yeasts in the feces of Mew gulls breeding in natural and urban habitats, with insights into the antifungal susceptibility of the observed pathogens. Birds (MDPI) 5(3):543–557 - https://doi.org/10.3390/birds5030036

The study focused on the comparison of culturable yeast communities in the feces of two populations of Mew Gulls depending on the breeding site: natural and urban. It was found that the species diversity of yeasts in the feces of gulls from the natural population was significantly higher, while the diversity of pathogenic species in the feces of birds from the anthropogenic population increased. The proportion of strains of pathogens (*Candida, Rhodotorula*) isolated from the feces of the anthropogenic colony that were resistant to conventional antibiotics was high. Indeed, populations of migratory birds breeding in an anthropogenic area could be involved in the global spread of the "alarming" geography of antifungal resistance, which is an urgent public health problem.

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Recent publications.

1 Sannino C, Qi W, Rüthi J, Stierli B, Frey B. 2023. Distinct taxonomic and functional profiles of high Arctic and alpine permafrost-affected soil microbiomes. Environ Microbiome 18(1):54 - https://doi.org/10.1186/s40793-023-00509-6

Background: Global warming is affecting all cold environments, including the European Alps and Arctic regions. Here, permafrost may be considered a unique ecosystem harboring a distinct microbiome. The frequent freeze– thaw cycles occurring in permafrost-affected soils, and mainly in the seasonally active top layers, modify microbial communities and consequently ecosystem processes. Although taxonomic responses of the microbiomes in permafrost-affected soils have been widely documented, studies about how the microbial genetic potential, especially pathways involved in C and N cycling, changes between active-layer soils and permafrost soils are rare. Here, we used shotgun metagenomics to analyze the microbial and functional diversity and the metabolic potential of permafrost-affected soil collected from an alpine site (Val Lavirun, Engadin area, Switzerland) and a High Arctic site (Station Nord, Villum Research Station, Greenland). The main goal was to discover the key genes abundant in the active-layer and permafrost soils, with the purpose to highlight the potential role of the functional genes found. **Results:** We observed differences between the alpine and High Arctic sites in alpha- and beta-diversity, and in EggNOG, CAZy, and NCyc datasets. In the High Arctic site, the metagenome in permafrost soil had an overrepresentation (relative to that in active-layer soil) of genes involved in lipid transport by fatty acid desaturate and ABC transporters, i.e. genes that are useful in preventing microorganisms from freezing by increasing membrane fluidity, and genes involved in cell defense mechanisms. The majority of CAZy and NCyc genes were overrepresented in permafrost soils relative to active-layer soils in both localities, with genes involved in the degradation of carbon substrates and in the degradation of N compounds indicating high microbial activity in permafrost in response to climate warming. **Conclusions:** Our study on the functional characteristics of permafrost microbiomes underlines the remarkably high functional gene diversity of the High Arctic and temperate mountain permafrost, including a broad range of C- and N-cycling genes, and multiple survival and energetic metabolisms. Their metabolic versatility in using organic materials from ancient soils undergoing microbial degradation determine organic matter decomposition and greenhouse gas emissions upon permafrost thawing. Attention to their functional genes is therefore essential to predict potential soil-climate feedbacks to the future warmer climate.

2 Bhunjun CS, Chen YJ, Phukhamsakda C, Boekhout T, Groenewald JZ, McKenzie EHC, Francisco EC, Frisvad JC, Groenewald M, Hurdeal VG, Luangsa-Ard J, Perrone G, Visagie CM, Bai FY, Błaszkowski J, Braun U, de Souza FA, de Queiroz MB, Dutta AK, Gonkhom D, Goto BT, Guarnaccia V, Hagen F, Houbraken J, Lachance MA, Li JJ, Luo KY, Magurno F, Mongkolsamrit S, Robert V, Roy N, Tibpromma S, Wanasinghe DN, Wang DQ, Wei DP, Zhao CL, Aiphuk W, Ajayi-Oyetunde O, Arantes TD, Araujo JC, Begerow D, Bakhshi M, Barbosa RN, Behrens FH, Bensch K, Bezerra JDP, Bilański P, Bradley CA, Bubner B, Burgess TI, Buyck B, Čadež N, Cai L, Calaça FJS, Campbell LJ, Chaverri P, Chen YY, Chethana KWT, Coetzee B, Costa MM, Chen Q, Custódio FA, Dai YC, Damm U, Santiago ALCMA, De Miccolis Angelini RM, Dijksterhuis J, Dissanayake AJ, Doilom M, Dong W, Álvarez-Duarte E, Fischer M, Gajanayake AJ, Gené J, Gomdola D, Gomes AAM, Hausner G, He MQ, Hou L, Iturrieta-González I, Jami F, Jankowiak R, Jayawardena RS, Kandemir H, Kiss L, Kobmoo N, Kowalski T, Landi L, Lin CG, Liu JK, Liu XB, Loizides M, Luangharn T, Maharachchikumbura SSN, Mkhwanazi GJM, Manawasinghe IS, Marin-Felix Y, McTaggart AR, Moreau PA, Morozova OV, Mostert L, Osiewacz HD, Pem D, Phookamsak R, Pollastro S, Pordel A, Poyntner C, Phillips AJL, Phonemany M, Promputtha I, Rathnayaka AR, Rodrigues AM, Romanazzi G, Rothmann L, Salgado-Salazar C, Sandoval-Denis M, Saupe SJ, Scholler M, Scott P, Shivas RG, Silar P, Silva-Filho AGS, Souza-Motta CM, Spies CFJ, Stchigel AM, Sterflinger K, Summerbell RC, Svetasheva TY, Takamatsu S, Theelen B, Theodoro RC, Thines M, Thongklang N, Torres R, Turchetti B, van den Brule T, Wang XW, Wartchow F, Welti S, Wijesinghe SN, Wu F, Xu R, Yang ZL, Yilmaz N, Yurkov A, Zhao L, Zhao RL, Zhou N, Hyde KD, Crous PW. 2024. What are the 100 most cited fungal genera? Stud Mycol. 108:1-411.

The global diversity of fungi has been estimated between 2 to 11 million species, of which only about 155 000 have been named. Most fungi are invisible to the unaided eye, but they represent a major component of biodiversity on our planet, and play essential ecological roles, supporting life as we know it. Although approximately 20 000 fungal genera are presently recognised, the ecology of most remains undetermined. Despite all this diversity, the mycological community actively researches some fungal genera more commonly than others. This poses an interesting question: why have some fungal genera impacted mycology and related fields more than others? To address this issue, we conducted a bibliometric analysis to identify the top 100 most cited fungal genera. A thorough database search of the Web of Science, Google Scholar, and PubMed was performed to establish which genera are most cited. The most cited 10 genera are *Saccharomyces*, *Candida*, *Aspergillus*, *Fusarium*, Penicillium, *Trichoderma*, *Botrytis*, *Pichia*, *Cryptococcus* and *Alternaria*. Case studies are presented for the 100 most cited genera with general background, notes on their ecology and economic significance and important research advances. This paper provides a historic overview of scientific research of these genera and the prospect for further research.

3 Canini F, Borruso L, Magrini S, D'Acqui LP, Buzzini P, Cavallini G, Zucconi L. 2024. Soil microbial communities dynamics in response to invasive groundcover *Carpobrotus*: insights into native species interactions and edaphic influence. Appl Soil Ecol 202:105537.

Alien invasive plant species of the genus *Carpobrotus* pose a major threat to coastal dune ecosystems. These human-introduced species have quickly expanded throughout the Mediterranean basin and other regions due to their high reproduction rates and adaptability. *Carpobrotus* invasion alters soil properties due to necromass and the release of allelopathic compounds, hindering the regrowth of native flora, with anticipated impacts on the native vegetation-associated soil microbial communities. While some studies have described changes in microbial communities between native and *Carpobrotus*-impacted areas, none have specifically addressed the responses of microbial communities associated with single native species. In this light, the bacterial and fungal communities specifically associated with different native species in both natural and *Carpobrotus*-impacted plots were examined in three locations along the middle Tyrrhenian Italian coast. Microbial communities responses to the *Carpobrotus* invasion varied greatly depending on the native species and the edaphic characteristics of the study locations. Microbial communities associated with *Pancratium maritimum* were the most affected ones and those associated with *Cakile maritima* the less sensitive to the invasion, which may be correlated to the different characteristics of these plant species. Furthermore, fungal communities exhibited a greater degree of disruption compared to bacterial ones. In invaded plots, fungal species that comprise plant pathogens were notably more abundant. This suggests that patterns in microbial communities response to this invasion phenomenon cannot be generalized, and that recovery strategies should consider various local conditions and be adjusted for the various native vegetation species. Finally, the possible spread of fungal plant pathogens as a mechanism of defense of *Carpobrotus* should be considered.

4 Piersanti S, Rebora M, Turchetti B, Salerno G, Ruscetta M, Zucconi L, D'Alò F, Buzzini P, Sannino C. 2024. Microplastics in the diet of *Hermetia illucens*: implications for development and midgut bacterial and fungal microbiota. Waste Management 186:259-270.

In a world with a population exceeding 8 billion people and continuing to grow, pollution from food and plastic waste is causing long-term issues in ecosystems. Potential solutions may be found by exploiting insect-based bioconversion. In this context, we investigated the impact of polyvinyl chloride microparticles (PVC-MPs) on the development of *Hermetia illucens* (black soldier fly; BSF) and its midgut bacterial and fungal microbiota. The impact of PVC-MPs was evaluated feeding BSF larvae with a PVC-MPs-supplemented diet. The larvae exposed to different PVC-MPs concentrations (2.5%, 5%, 10% and 20% w/w) developed into adults with no significant increase in pupal mortality. Faster development and smaller pupae were observed when 20% PVCMPs was provided. The BSF larvae ingest PVC-MPs, resulting in a reduction in MPs size. Larvae exposed to PVC-MPs did not exhibit differences in gut morphology. Regarding the impact of PVC-MPs on the structure of both bacterial and fungal communities, the overall alpha- and beta-diversity did not exhibit significant changes. However, the presence of PVC-MPs significantly affected the relative abundances of Enterobacteriaceae and Paenibacillaceae among the bacteria and of Dipodascaceae and Plectospharellaceae among the fungi (including yeast and filamentous life forms), suggesting that PVC-MP contamination has a taxa-dependent impact. These results indicate that BSF larvae can tolerate PVC-MPs in their diet, supporting the potential use of these insects in organic waste management, even in the presence of high levels of PVC-MP contamination.

5 Mugnai G, Pinchuk I, Borruso L, Tiziani R, Sannino C, Canini F, Turchetti B, Mimmo T, Zucconi L, Buzzini P. 2024. The hidden network of biocrust successional stages in the High Arctic: revealing abiotic and biotic factors shaping microbial and metazoan communities. Sci Total Environ 926:171786.

Despite the important role that biocrust communities play in maintaining ecosystem structure and functioning in deglaciated barren soil, few studies have been conducted on the dynamics of biotic communities and the impact of physicochemical characteristics in shaping the different successional stages. In this study an integrated approach encompassing physicochemical parameters and molecular taxonomy was used for identifying the indicator taxa and the presence of intra- and inter-kingdom interactions in five different crust/biocrust successional stages: i) physical crust, ii) cyanobacteria-dominated biocrust, iii) cyanobacteria/moss-dominated biocrust, iv) moss-dominated biocrust and v) bryophyte carpet. The phylum Gemmatimonadota was the bacterial indicator taxon in the early stage, promoting both inter- and intra-kingdom interactions, while Cyanobacteria and Nematoda phyla played a pivotal role in formation and dynamics of cyanobacteria-dominated biocrusts. A multitrophic community, characterized by a shift from oligotrophic to copiotrophic bacteria and the presence of saproxylic arthropod and herbivore insects was found in the cyanobacteria/moss-dominated biocrust, while a more complex biota, characterized by an increased fungal abundance (classes Sordariomycetes, Leotio-mycetes, and Dothideomycetes, phylum Ascomycota), associated with highly trophic consumer invertebrates (phyla Arthropoda, Rotifera, Tardigrada), was observed in moss-dominated biocrusts. The class Bdelloidea and the family Hypsibiidae (phyla Rotifera and Tardigrada, respectively) were metazoan indicator taxon in bryophyte carpet, suggesting their potential role in shaping structure and function of this late successional stage. Nitrogen and phosphorus were the main physicochemical limiting factors driving the shift among different crust/biocrust successional stages. Identification and characterization of indicator taxa, biological intra- and inter-kingdom interactions and abiotic factors driving the shift among different crust/biocrust successional stages provide a detailed picture on crust/biocrust dynamics, revealing a strong interconnection among micro- and macrobiota systems. These findings enhance our understanding of biocrust ecosystems in High Arctic, providing valuable insights for their conservation and management in response to environmental shifts due to climate change.

6 Fracasso I, Zaccone C, Oskolkov N, Da Ros L, Dinella A, Belelli Marchesini L, Buzzini P, Sannino C, Turchetti B, Cesco S, Le Roux G, Tonon G, Vernesi C, Mimmo T, Ventura M, Borruso L. 2024. Exploring different methodological approaches to unlock paleobiodiversity in peat profiles using ancient DNA. Sci Total Environ 908:168159.

Natural and human-induced environmental changes deeply affected terrestrial ecosystems throughout the Holocene. Paleoenvironmental reconstructions provide information about the past and allow us to predict/model future scenarios. Among potential records, peat bogs are widely used because they present a precise stratigraphy and act as natural archives of highly diverse organic remains. Over the decades, several techniques have been developed to identify debris occurring in peat, including their morphological description. However, this is strongly constrained by the researcher's ability to distinguish residues at the species level, which typically requires many years of experience. In addition, potential contamination hampers using these techniques to obtain information from organisms such as fungi or bacteria. Environmental DNA metabarcoding and shotgun metagenome sequencing could represent a solution to detect specific groups of organisms without any a priori knowledge of their characteristics and/or to identify organisms that have rarely been considered in previous investigations. Moreover, shotgun metagenomics may allow the identification of bacteria and fungi (including both yeast and filamentous life forms), ensuring discrimination between ancient and modern organisms through the study of deamination/damage patterns. In the present review, we aim to i) present the state-of-the-art methodologies in paleoecological and paleoclimatic studies focusing on peat core analyses, proposing alternative approaches to the classical morphological identification of plant residues, and ii) suggest biomolecular approaches that will allow the use of proxies such as invertebrates, fungi, and bacteria, which are rarely employed in paleoenvironmental reconstructions.

7 Leo P, Onofri S, Zucconi L, Selbmann L, Turchetti B, Buzzini P, Chander AM, Simpson A, Singh N, Vellone D, Tighe S, Venkateswaran K. 2023. Draft genome sequencing of *Naganishia* species isolated from the polar environments. Microbiol Resour Announc 12:e00388-23.

The draft genomes of five *Naganishia* strains were sequenced using MinION and annotated using Funannotate pipeline. Phylogenetic and genomic analyses were performed to provide their genetic relationships, diversity, and potential functional capabilities. This approach will aid in understanding their potential to survive under microgravity and their resilience to extreme environments.

8 Turchetti B, De Francesco G, Mugnai G, Sileoni V, Alfeo V, Buzzini P, Yurkov A, Marconi O. 2023. Species and temperature-dependent fermentative aptitudes of *Mrakia* genus for innovative brewing. Food Res Int 170:113004.

The use of non-conventional brewing yeasts as alternative starters is a very promising approach which received increasing attention from worldwide scientists and brewers. Despite the feasible application of non-conventional yeasts in brewing processes, their regulations and safety assessment by the European Food Safety Authority still represent a bottlenecked hampering their commercial release, at least into EU market. Thus, research on yeast physiology, accurate taxonomic species identification and safety concerns associated with the use of nonconventional yeasts in food chains is needed to develop novel healthier and safer beers. Currently, most of the documented brewing applications catalyzed by non-conventional yeasts are associated to ascomycetous yeasts, while little is known about analogous uses of basidiomycetous taxa. Therefore, in order to extend the phenotypic diversity of basidiomycetous brewing yeasts the aim of this investigation is to check the fermentation aptitudes of thirteen *Mrakia* species in relation to their taxonomic position within the genus *Mrakia*. The volatile profile, ethanol content and sugar consumption were compared with that produced by a commercial starter for low alcohol beers, namely *Saccharomycodes ludwigii* WSL 17. The phylogeny of *Mrakia* genus showed three clusters that clearly exhibited different fermentation aptitudes. Members of *M. gelida* cluster showed a superior aptitude to produce ethanol, higher alcohols, esters and sugars conversion compared to the members of *M. cryoconiti* and *M. aquatica* clusters. Among *M. gelida* cluster, the strain *M. blollopis* DBVPG 4974 exhibited a medium flocculation profile, a high tolerance to ethanol and to iso-α-acids, and a considerable production of lactic and acetic acids, and glycerol. In addition, an inverse relationship between fermentative performances and incubation temperature is also displayed by this strain. Possible speculations on the association between the cold adaptation exhibited by *M. blollopis* DBVPG 4974 and the release of ethanol in the intracellular matrix and in the bordering environment are presented.

9 Turchetti B, Bevivino A, Casella P, Coleine C, Felis GE, Girometta CE, Molino A, Perugini I, Pollio A, Prigione V, Selbmann L, Varese C, Buzzini P. 2023. Selected case studies on fastidious eukaryotic microorganisms: issues and investigation strategies. Diversity 15:862.

The concept of fastidious microorganisms currently found in scientific literature is mainly related to the difficulty of isolating/culturing/preserving bacteria. Eukaryotes are investigated much less in this respect, although they represent a fundamental part of the microbial world. Furthermore, not only isolation, but also identification and culturing (in the perspective of long-term preservation) should be considered key aspects often impacting on the study of fastidious microorganisms, especially in terms of preservation in culture collections and biotechnological exploitation. The present review aimed to investigate the current state of the art on fastidious eukaryotes, with special emphasis on the efforts to improve their isolation, identification, culturing and long-term preservation in culture collections practices. A few case studies focused on some fastidious eukaryotic microorganisms (including possible customized solutions to overcome specific issues) are also presented: isolation and preservation of slow-growing fungi, culturing of *Haematococcus lacustris*, isolation of unialgal strains of Cyanidiophytina (Rhodophyta), identification of *Metschnikowia pulcherrima* clade yeasts, isolation and preservation of *Pyricularia* species, preservation of *Halophytophtora* spp.

10 Biagioli F, Coleine C, Buzzini P, Turchetti B, Sannino C, Selbmann L. 2023. Positive fungal interactions are key drivers in Antarctic endolithic microcosms at the boundaries for life sustainability. FEMS Microbiol Ecol 99:1-8.

In the ice-free areas of Victoria Land in continental Antarctica, where the conditions reach the limits for life sustainability, highly adapted and extreme-tolerant microbial communities exploit the last habitable niches inside porous rocks (i.e. cryptoendolithic communities). These guilds host the main standing biomass and principal, if not sole, contributors to environmental/biogeochemical cycles, driving ecosystem processes and functionality in these otherwise dead lands. Although knowledge advances on their composition, ecology, genomic and metabolic features, a large-scale perspective of occurring interactions and interconnections within and between endolithic fungal assemblages is still lacking to date. Unravelling the tight relational network among functional guilds in the Antarctic cryptoendolithic communities may represent a main task. Aiming to fill this knowledge gap, we performed a correlationnetwork analysis based on amplicon-sequencing data of 74 endolithic microbiomes collected throughout Victoria Land. Endolithic communities' compositional pattern was largely dominated by Lichenized fungi group (83.5%), mainly represented by Lecanorales and Lecideales, followed by Saprotrophs (14.2%) and RIF+BY (2.4%) guilds led by Tremellales and Capnodiales respectively. Our findings highlighted that fungal functional guilds' relational spectrum was dominated by cooperative interactions led by lichenised and black fungi, deeply engaged in community trophic sustain and protection, respectively. On the other hand, a few negative correlations found may help in preserving niche boundaries between microbes living in such strict spatial association.

11 Isola D, Prigione V, Zucconi L, Varese GC, Poli A, Turchetti B, Canini F. 2022. *Knufia obscura* sp. nov. and *Knufia victoriae* sp. nov., two new species from extreme environments. Int J Syst Evol Microbiol 72:005530.

Six strains of black meristematic fungi were isolated from Antarctic soils, gasoline car tanks and from the marine alga *Flabellia petiolata*. These fungi were characterized by morphological, physiological and phylogenetic analyses. According to the maximum-likelihood analysis reconstructed with ITS and LSU sequences, these strains belonged to the genus *Knufia. Knufia obscura* sp. nov. (holotype CBS 148926) and *Knufia victoriae* sp. nov. (holotype CBS 149015) are proposed as two novel species and descriptions of their morphological, physiological and phylogenetic features are presented. Based on the maximum-likelihood analyses, *K. obscura* was closely related to *Knufia hypolithi* (99 % bootstrap support), while *K. victoriae* clustered in the clade of *Knufia cryptophialidica* and *Knufia perfecta* (93 % bootstrap support). *Knufia victoriae*, recorded in Antarctic soil samples, had a psychrophilic behaviour, with optimal growth between 10 and 15 °C and no growth recorded at 20 °C. *Knufia obscura*, from a gasoline car tank and algae, displayed optimal growth between 20 and 25 °C and was more tolerant to salinity than *K. victoriae*.

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Recent publication.

1 Peri KVR, Yuan L. Oliveira FF, Persson K, Alalam HD, Olsson L, Larsbrink J, Kerkhoven EJ, Geijer C. 2004. A unique metabolic gene cluster regulates lactose and galactose metabolism in the yeast *Candida intermedia.* Appl Environ Microbiol 90(10):e01135-24 - https://doi.org/10.1128/aem.01135-24

Lactose assimilation is a relatively rare trait in yeasts, and *Kluyveromyces* yeast species have long served as model organisms for studying lactose metabolism. Meanwhile, the metabolic strategies of most other lactose-assimilating yeasts remain unknown. In this work, we have elucidated the genetic determinants of the superior lactose-growing yeast *Candida intermedia*. Through genomic and transcriptomic analyses, we identified three interdependent gene clusters responsible for the metabolism of lactose and its hydrolysis product galactose: the conserved *LAC* cluster (*LAC12*, *LAC4*) for lactose uptake and hydrolysis, the conserved *GAL* cluster (*GAL1*, *GAL7*, and *GAL10*) for galactose catabolism through the Leloir pathway, and a "*GALLAC*" cluster containing the transcriptional activator gene *LAC9*, second copies of *GAL1* and *GAL10*, and a *XYL1* gene encoding an aldose reductase involved in carbon overflow metabolism. Bioinformatic analysis suggests that the *GALLAC* cluster is unique to *C. intermedia* and has evolved through gene duplication and divergence, and deletion mutant phenotyping proved that the cluster is indispensable for *C. intermedia's* growth on lactose and galactose. We also show that the regulatory network in *C. intermedia*, governed by Lac9 and Gal1 from the *GALLAC* cluster, differs significantly from the galactose and lactose regulons in *Saccharomyces cerevisiae*, *Kluyveromyces lactis*, and *Candida albicans*. Moreover, although lactose and galactose metabolism are closely linked in *C. intermedia*, our results also point to important regulatory differences.

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Recent publication.

1 Rij M, Kayacan Y, Bernardi B, Wendland J. 2024. Re-routing MAP kinase signaling for penetration peg formation in predator yeasts. PLoS pathogens. 20(8):e1012503 - https://doi.org/10.1371/journal.ppat.1012503

Saccharomycopsis yeasts are natural organic sulfur auxotrophs due to lack of genes required for the uptake and assimilation of sulfate/sulfite. Starvation for methionine induces a shift to a predatory, mycoparasitic life strategy that is unique amongst ascomycetous yeasts. Similar to fungal plant pathogens that separated from *Saccharomycopsis* more than 400 million years ago, a specialized infection structure called penetration peg is used for prey cell invasion. Penetration pegs are highly enriched with chitin. Here we demonstrate that an ancient and conserved MAP kinase signaling pathway regulates penetration peg formation and successful predation in the predator yeast *S. schoenii*. Deletion of the MAP kinase gene SsKIL1, a homolog of the *Saccharomyces cerevisiae* ScKSS1/ScFUS3 and the rice blast Magnaporthe oryzae MoPMK1 genes, as well as deletion of the transcription factor SsSTE12 generate non-pathogenic mutants that fail to form penetration pegs. Comparative global transcriptome analyses using RNAseq indicate loss of the SsKil1-SsSte12-dependent predation response in the mutant strains, while a methionine starvation response is still executed. Within the promoter sequences of genes upregulated during predation we identified a cis-regulatory element similar to the ScSte12 pheromone response element. Our results indicate that, re-routing MAP-kinase signaling by re-wiring Ste12 transcriptional control towards predation specific genes contributed to the parallel evolution of this predacious behaviour in predator yeasts. Consequently, we found that SsSTE12 is dispensable for mating.

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The following papers, authored by PYCC members were recently published.

1 Avchar R, Giri G, Thakkar L, Sharma A, Sampaio JP. 2024. *Wickerhamiella lachancei* f.a. sp. nov. a novel ascomycetous yeast species isolated from flowers of *Lantana camara* in India. Int J Syst Evol Microbiol 74:1466-5034. https://doi.org/10.1099/ijsem.0.006365

2 Sakpuntoon V, Srathongporn N, Pontes A, Khunnamwong P, Aires A, Limtong S, Gonçalves C, Gonçalves P, Sampaio JP, Srisuk N. 2024. Phylogenomic delineation of two new species of ascomycetous yeasts, *Wickerhamiella koratensis* sp. nov. and *Wickerhamiella limtongiae* sp. nov., and proposal of two synonyms, *Wickerhamiella infanticola* and *Wickerhamiella tropicalis*. Int J Syst Evol Microbiol 74(2):006282 - https://doi.org/10.1099/ijsem.0.006282

3 Pontes A, Paraíso F, Silva M, Lagoas C, Aires A, Brito PH, Rosa CA, Lachance M-A, Sampaio JP, Gonçalves C, Gonçalves P. 2024. Extensive remodeling of sugar metabolism through gene loss and horizontal gene transfer in a eukaryotic lineage. BMC Biology 22:128 - https://doi.org/10.1186/s12915-024-01929-7

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

Recent publications.

1 Jiménez-Padilla Y, Chan Y, Aletta MS, Lachance MA, Simon AF. 2024. The effect of microbiome on social spacing in *Drosophila melanogaster* depends on genetic background and sex. Micropublication Biology - https://doi.org/10.17912/micropub.biology.001270

The gut microbiome modulates many essential functions including metabolism, immunity, and behaviour. Specifically, within behaviour, social behaviours such as sociability, aggregation, mating preference, avoidance, oviposition, and aggression are known to be regulated in part by this host-microbiome relationship. Here, we show the microbiome's role in the determination of social spacing in a sex- and genotype-specific manner. Future work can be done on characterizing the microbiome in each of these fly strains to identify the species of microbes present as well as their abundance.

2 Barros KO, Valério AD, Batista TM, Santos AR, Souza GF, Alvarenga FB, Lopes MR, Morais CG, Alves C, Goes-Neto A, Vital MJ, Uetanabaro APT, Souza DT, Bai FY, Franco GR, Lachance MA, Rosa CA, Johann S. 2024. *Spencermartinsiella nicolii* sp. nov., a potential opportunistic pathogenic yeast species isolated from rotting wood in Brazil. Int J Syst Evol Microbiol. 74(9):006520 - https://doi.org/10.1099/ijsem.0.006520

Nineteen isolates representing a candidate for a novel yeast species belonging to the genus Spencermartinsiella were recovered from rotting wood samples collected at different sites in Atlantic Rainforest and Amazonian Forest ecosystems in Brazil. Similarity search of the nucleotide sequence of the intergenic spacer (ITS)-5.8S and large subunit D1/D2 regions of the ribosomal gene cluster showed that this novel yeast is closely related to *Spencermartinsiella cellulosicola*. The isolates differ by four nucleotide substitutions in the D1/D2 domain and six substitutions and 31 indels in the ITS region from the holotype of S. cellulosicola. Phylogenomic analysis based on 1474 single-copy orthologues for a set of *Spencermartinsiella* species whose whole genome sequences are available confirmed that the novel species is phylogenetically close to S. cellulosicola. The low average nucleotide identity value of 83% observed between *S. cellulosicola* and the candidate species confirms that they are distinct. The novel species produced asci with hemispherical ascospores. The name *Spencermartinsiella nicolii* sp. nov. is proposed. The holotype is CBS 14238T. The MycoBank number is MB855027. Interestingly, the D1/D2 sequence of the S. nicolii was identical to that of an uncultured strain of *Spencermartinsiella* causing systemic infection in a male adult crocodile (*Crocodylus niloticus*). The characterization of some virulence factors and antifungal susceptibility of *S. nicolii* isolates suggest that this yeast may be an opportunistic pathogen for animals, including humans; the isolates grow at 37 °C.

3 García-Acero AM, Batista TM, Souza GFL, Santos ARO, Souza DL, Franco GR, Velásquez-Lozano ME Yamamoto D, Toki W, Lachance MA, Rosa CA. 2024. Description of Millerago gen. nov. based on taxogenomic analysis, with two new species, *Millerago phaffii* f.a., sp. nov., and *Millerago galiae* f. a., sp. nov. Int J Syst Evol Microbiol 74(10):006565 - https://doi.org/10.1099/ijsem.0.006565

Four yeast isolates obtained from tree bark and fermenting sap of *Quercus* spp. and insects in Colombia and Japan were phylogenetically related to *Candida galis* based on analyses of the sequences of the internal transcribed spacer (ITS) region and the D1/D2 domains of the large subunit rRNA gene. The novel species differs from *C. galis* by 20 nt substitutions and 5 indels in the D1/D2 sequences. A phylogenomic analysis suggested that these species are related to *Candida ficus*, the genus *Phaffomyces* and a small clade containing *Barnettozyma botsteinii*, *Barnettozyma siamensis* and *Candida montana*. Our genomic analyses suggest that the novel species and *C. galis* should be separated in a novel yeast genus. We propose the genus *Millerago* gen. nov. to accommodate these species and the species *Millerago phaffii* f.a., sp. nov. (CBS 18021T; MycoBank MB856172) to accommodate the Colombian and Japanese isolates. The Colombian isolate of *M. phaffii* differs from the Japanese isolates by three nt substitutions and one indel and two substitutions and one indel in the ITS and D1/D2 sequences, respectively, showing that they were conspecific. We also propose the new species *Millerago galiae* sp. nov. to validate this species according to the rules of the International Code of Nomenclature for algae, fungi and plants.

4 Santa-Brigída R, Santos ARO, Martins MB, Rosa LH, Lachance MA, Rosa CA. 2024. *Kodamaea schenbergiae* f.a., sp. nov. and *Suhomyces schwaniae* f.a., sp. nov., two yeast species isolated from mushrooms and associated insects in a Brazilian Amazonian rainforest biome. Int J Syst Evol Microbiol (accepted November 2024).

5 Yurkov A, Visagie CM, Crous PW, Hashimoto A, Baschien C, Begerow D, Kemler M, Schoutteten N, Stadler M, Wijayawardene NN, Hyde KD, Zhang N, Boekhout T, ICTF Yeast Working Group*, May TW, Thines M, Hawksworth DL. ICTF Yeast Working Group: Yurkov A, Boekhout T, Bai FY, Begerow D, Čadež N, Daniel HM, Fell JW, Groenewald M, Lachance MA, Libkind D, Péter G, Takashima M, Turchetti B. 2024. Cultures as types and the utility of viable specimens for fungal nomenclature. IMA Fungus 15(1):20 - https://doi.org/10.1186/s43008-024-00155-8

The debates over the requirement of the International Code of Nomenclature for algae, fungi, and plants (ICNafp) for a viable specimen to represent the name-bearing type material for a species or infraspecific taxon have a long history. Taxonomy of fungi commonly studied as living cultures exemplified by yeasts and moulds, strongly depend on viable reference material. The availability of viable cultures is also particularly useful for several groups of filamentous and dimorphic fungi. While the preservation of metabolically inactive cultures is permitted and recommended by the ICNafp, there is room for improvement. Below, we review the history and current status of cultures as the name-bearing type material under the Code. We also present a roadmap with tasks to be achieved in order to establish a stable nomenclatural system that properly manages taxa typified by viable specimens. Furthermore, we propose setting up rules and defining the nomenclatural status of ex-type cultures under Chapter F, the section of the ICNafp that includes provisions specific to names of fungi.

XIV National Institutes of Science and Technology, Brazil. Communicated by A.K. Gombert <gombert@unicamp.br>.

The following articles were published under the framework of INCT Yeast (National Institutes of Science and Technology), a research network based in Brazil, involving 11 Brazilian universities and seven international universities, and coordinated by Prof. Carlos Rosa. The aim is to prospect, characterize, and preserve new yeast strains, and to develop biotechnological innovation.

1 Tadioto V, Giehl A, Cadamuro RD, Guterres IZ, dos Santos AA, Bressan SK, Werlang L, Stambuk BU, Fongaro G, Silva IT, Alves SL Jr. 2023. Bioactive Compounds from and against Yeasts in the One Health Context: A Comprehensive Review. Fermentation (MDPI) *9*(4):363 - https://doi.org/10.3390/fermentation9040363

2 Santos ARO, Barros KO, Batista TM, Souza GFL, Alvarenga FBM, Abegg MA, Sato TK, Hittinger CT, Lachance MA, Rosa CA. 2023. *Saccharomycopsis praedatoria* sp. nov, a predacious yeast isolated from soil and rotten wood in an Amazonian rainforest biome. Int J Syst Evol Microbiol 73(10):006125 - https://doi.org/10.1099/ijsem.0.006125

3 Souza GFL, Barros KO, Alvarenga FBM, Santos ARO, Fonseca CRV, Abegg MA, Lachance MA, Rosa CA. 2023. *Sugiyamaella bielyi* f. a, sp. nov. and *Sugiyamaella amazoniana* f. a, sp. nov, two yeast species isolated from passalid beetles and rotting wood in Amazonia. Int J Syst Evol Microbiol 73(4):005839 - https://doi.org/10.1099/ijsem.0.005839

4 Albarello MLR, Giehl A, Tadioto V *et al.* 2023. Analysis of the holocellulolytic and fermentative potentials of yeasts isolated from the gut of *Spodoptera frugiperda* Larvae. Bioenerg Res 16:2046–2057 - https://doi.org/10.1007/s12155-023-10616-4

5 Giehl A, dos Santos AA, Cadamuro RD, Tadioto V, Guterres IZ, Prá Zuchi ID, Minussi GdA, Fongaro G, Silva IT, Alves SLJr. 2023. Biochemical and biotechnological insights into fungus-plant interactions for enhanced sustainable agricultural and industrial processes. Plants (MDPI) 12(14):2688 - https://doi.org/10.3390/plants12142688

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9 Palladino F, Alvarenga FBM, de Cássia Lacerda Brambilla Rodrigues R. *et al.* 2023. Diversity of native yeasts isolated in Brazil and their biotechnological potential for the food industry. Curr Food Sci Tech Rep 1:81–90 - https://doi.org/10.1007/s43555-023-00011-7

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26 García-Acero AM, Batista TM, Souza GFL, Santos ARO, Souza DL, Franco GR, Velásquez-Lozano ME, Yamamoto D, Toki W, Lachance MA, Rosa CA. 2024. Description of *Millerago* gen. nov. based on taxogenomic analysis, with two new species, *Millerago phaffii* f.a, sp. nov. and *Millerago galiae* f.a, sp. nov. Int J Syst Evol Microbiol 74(10):006565 - https://doi.org/10.1099/ijsem.0.006565

Forthcoming Meetings

Please save the date for the 32nd edition of the International Conference on Yeast Genetics and Molecular Biology (ICYGMB) which will take place on July 21-24, 2025 at the International Conference Centre of Sorbonne University in Paris, France.

Check out the conference website https://premc.org/yeast2025/ for information on the program, the lineup of invited speakers and much more. For further information, email yeast2025@premc.org.

38th International Specialized Symposium on Yeasts (ISSY38)
Warsaw, Poland, 1-5 September 2025

On behalf of the International Commission on Yeasts, the Local Conference Organizing Committee, and the International Scientific Committee, it is our great pleasure to invite you to the 38th International Specialized Symposium on Yeasts (ISSY38). The symposium will be held as an in-person event from 1 to 5 September 2025 in Warsaw, Poland.

It is the first time in the symposium's 60-year history that it will be held in Poland. The symposium is jointly organized by the International Commission on Yeasts (ICY), the Institute of Biochemistry and Biophysics of the Polish Academy of Sciences, the Wroclaw University of Environmental and Life Sciences, and the Biology Department of the University of Warsaw. The venue of ISSY38 will be at the beautiful historical campus of University of Warsaw.

The theme of the ISSY38 conference is Yeast, the Omni-Tool. This theme will bring together a broad cross section of investigators working on various yeast species, researchers in basic science and industry scientists, and students, allowing them to share their latest discoveries. It will facilitate and encourage communication between the international yeast society and Polish yeast researcher communities, as many Polish universities and institutes have strong and rapidly growing research teams studying fundamental and applied aspects of yeast biology and biotechnology.

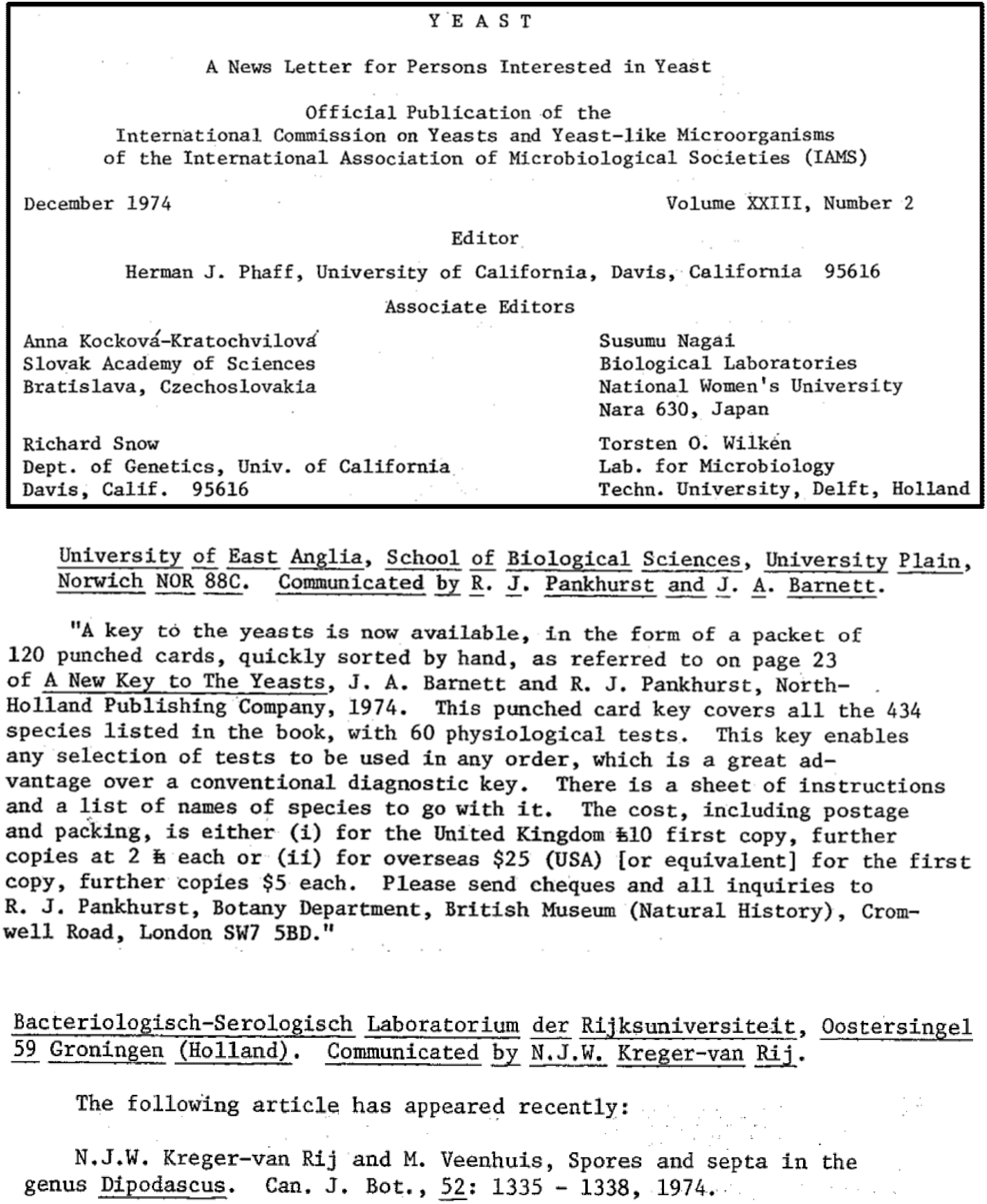
The main topics of ISSY38 will be:

- Deciphering of molecular mechanisms of eukaryotic cells with yeast
- Yeast screens
- Metabolism and metabolomics
- Yeasts as molecular factories
- Fermentation, food and beverages
- Yeast in population and evolution models
- Humanized yeast in health research

We look forward to seeing you in Warsaw! Registration is opened. For more information: https://issy38.com.pl/

Adrianna Skoneczna and Zbigniew Lazar, Co-chairs of ISSY38

Fifty Years Ago



YEAST

A News Letter for Persons Interested in Yeast

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University of East Anglia, School of Biological Sciences, University Plain, Norwich NOR 88C. Communicated by R. J. Pankhurst and J. A. Barnett.

"A key to the yeasts is now available, in the form of a packet of 120 punched cards, quickly sorted by hand, as referred to on page 23 of A New Key to The Yeasts, J. A. Barnett and R. J. Pankhurst, North-Holland Publishing Company, 1974. This punched card key covers all the 434 species listed in the book, with 60 physiological tests. This key enables any selection of tests to be used in any order, which is a great advantage over a conventional diagnostic key. There is a sheet of instructions and a list of names of species to go with it. The cost, including postage and packing, is either (i) for the United Kingdom £10 first copy, further copies at 2 £ each or (ii) for overseas $25 (USA) [or equivalent] for the first copy, further copies $5 each. Please send cheques and all inquiries to R. J. Pankhurst, Botany Department, British Museum (Natural History), Cromwell Road, London SW7 5BD."

Bacteriologisch-Serologisch Laboratorium der Rijksuniversiteit, Oostersingel 59 Groningen (Holland). Communicated by N.J.W. Kreger-van Rij.

The following article has appeared recently:

N.J.W. Kreger-van Rij and M. Veenhuis, Spores and septa in the genus Dipodascus. Can. J. Bot., 52: 1335 - 1338, 1974.

National Collection of Yeast Cultures, Lyttel Hall, Nutfield, Redhill, Surrey RH1 4HY, England. Communicated by Barbara Kirsop, Curator, N.C.Y.C.

The NCYC now contains over 1050 different yeast strains. They are at present maintained in liquid MYGP medium at 4°C and subcultured every 6 months. Between 1954 and 1960 many of the strains in the collection were also freeze-dried by the method described in the Journal of the Institute of Brewing, 1955, 61, 566.

During 1972-73 all the yeasts maintained in the NCYC, apart from some recently deposited strains and some genetically marked mutants, were examined morphologically and biochemically. Appropriately significant tests were selected for each strain and results were compared with the original descriptions at the time of deposition. It was found that a significant amount of variation had taken place in both the biochemical and morphological properties of strains that had been maintained conventionally for long periods. Equivalent freeze-dried cultures of many of the yeasts were compared with the subcultured strains and it was found that properties of the lyophilized cultures had remained stable. Although initial viability of cultures immediately after freeze-drying had been disappointing, only a very small subsequent decrease in viability has been detected after storage for between 10 and 20 years. Detailed study of brewing yeasts showed that brewing performance remained unchanged after prolonged storage in the freeze-dried state.

Since it is considered that the major function of a culture collection is to maintain cultures in such a way that they represent accurately the strains that were deposited, it is now the intention to lyophilize the majority of the strains held in the NCYC. Strains that have been shown to react poorly to lyophilization will continue to be maintained conventionally and will also be maintained experimentally on silica gel.

Zoology Department, West Mains Road, Edinburgh University, EH9-3JT, U.K.. Communicated by J. M. Mitchison.

Our present group working on the yeast cell cycle (mainly Schizosaccharomyces pombe) includes Dr. Bruce Carter, Jim Creanor, Dr. Peter Fantes, Davor Fatori, Kim Nasymth, Dr. Paul Nurse, Dr. Chris Sissons and Bill Staatz. Dr. John May from Monash University will be here until the end of the year. Next year, we are expecting Dr. Alan Day from the University of Western Ontario, and Dr. Pierre Thuriaux and Dr. Michele Thuriaux-Minet from Bern University.

University of Amsterdam, Section for Medical Enzymology, Laboratory of Biochemistry, Eerste Constantijn Huygens Straat 20, Amsterdam, The Netherlands. Communicated by G.S.P. Groot.

SEQUENCE HOMOLOGY OF NUCLEAR AND MITOCHONDRIAL DNAs OF DIFFERENT YEASTS.

G.S.P. Groot, R. A. Flavell, and J.P.M. Sanders

Biochimica et Biophysica Acta, in press

SUMMARY

1. Both nuclear and mtDNA of four different yeasts show approximately 10% homology as measured by DNA-DNA filter hybridization. These homologous sequences are mainly attributable to the ribosomal cistrons.

2. Melting curve analysis shows that the heterologous mitochondrial DNA-DNA hybrids contain several times more mismatching than the nuclear DNA-DNA hybrids.

3. DNA-rRNA hybridization shows that the sequences of the ribosomal cistrons in both the nuclear and the mitochondrial genome have been conserved during evolution.

4. However, melting curve analysis of the DNA-rDNA hybrids shows that the sequence of the nuclear ribosomal cistrons have undergone considerably fewer nucleotide substitutions than their mitochondrial counterparts.

5. The results suggest that the mitochondrial ribosomal cistrons have evolved more rapidly than the nuclear cistrons. This is discussed in the light of theories on the rate of molecular evolution.

Brandeis University, Rosenstiel Basic Medical Sciences Research Center, Waltham Massachusetts 02154. Communicated by James E. Haber.

Bisexual Mating Behavior in a Diploid of *Saccharomyces cerevisiae*: Evidence for Genetically Controlled Non-Random Chromosome Loss During Vegetative Growth.

The Research Laboratories of Kirin Brewery Co., Ltd. Takasaki, Gumma Pref., Japan. Communicated by Tatsuhiko Kaneko.

The following is the abstract of a paper presented at the First Intersectional Congress of the International Association of Microbiological Societies, Tokyo, Japan, September 1-7, 1974.

YEAST PROTOPLAST FORMATION BY A PURIFIED ENZYME, ZYMOLYASE, AND ITS REGENERATION

Kumpei Kitamura and Yasushi Yamamoto

Protoplasts were prepared from cells of Sacch. carlsbergensis by Zymolyase, which is a purified glucanase from Arthrobacter luteus and hydrolyzes β-1, 3-glucan, specifically releasing laminaripentaose as the minimum product. The yeast cells cultivated in a yeast extract medium containing glucose were treated by Zymolyase in a reaction mixture containing 0.6 M KCl and 0.1 M $MgSO4$ as a stabilizer at 25°C with gentle shaking. After incubation for 10 min, destruction began to be observed sporadically over the cell wall, and then protoplasm became spherical within the perforated and worn envelope, and, at last the protoplast slipped out the remaining envelope faintly visible. When the protoplasts were incubated in a liquid medium containing 0.6 M KCl or 0.8 M sucrose at 30°C, the protoplasts began to grow within 1 hr and then they changed to a variety of forms to show increase of cellular materials. After 24 hr, regeneration of normal cells from the protoplasts was seen to occur in a medium containing sucrose.

Amoco Foods Company, Amoco Research Center, Naperville, Illinois 60540, U.S.A. Communicated by C. Akin and R. J. Flannery

Torula Yeast from Ethanol: The First Large Scale Production of Food From A Non-Agricultural Source:

Standard Oil Company of Indiana, after a nine-year multi-million dollar research program, has announced a new process to grow Torula Yeast on ethyl alcohol. Standard's subsidiary, Amoco Foods Company, is building at Hutchinson, Minn., its first commercial plant with a capacity of more than 10 million pounds of yeast a year. The Amoco Torula Yeast (Candida utilis) is grown in an aseptic continuous fermentor, and harvested under sanitary conditions. This food yeast is over 50 percent protein and contains vitamins and minerals, and meets Food and Drug Administration regulations.

Amoco Foods will market the yeast to independent food processors who will incorporate the high protein ingredient in their food products in order to improve nutrition, flavor, cost or other properties of their processed foods.
