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YEAST

A Newsletter for Persons Interested in Yeast

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Foreign Subscribers: It has come to our attention that mailing of the Yeast Newsletter by printed matter involves a 2-3 month delay in your receiving it. If you are not receiving the Yeast Newsletter by airmail (which takes approximately 2 weeks) and would like to, please let us know. An additional $4 per year is required to cover postage and handling for this service.

I wish all readers of the Yeast Newsletter a prosperous and scientifically rewarding New Year.

Herman J. Phaff
Editor

I. American Type Culture Collection, 12301 Parklawn Drive, Rockville, Maryland 20852. Communicated by S.C. Jong.

The strains listed below have been added to the ATCC since May 4, 1981. Complete information for these strains may be obtained upon request from the Mycology Department of ATCC.

Yarrowia lipolytica ATCC 44601 produces citrate	Dr. Aiba Osaka University Japan
Schizosaccharomyces pombe ATCC 44608 - 44611, 44625 - 44634 genetic	P. Munz University of Bern Switzerland
Schizosaccharomyces pombe ATCC 44635 genetic	P. Thuriaux University of Bern Switzerland
Rhodotorula ingeniosa ATCC 44636 assimilation of methanol	W. Babel E. Germany
Candida boidinii assisilation of methanol	"
Candida utilis ATCC 44638 - 44642 sensitive to antimycin	Y. Iwamoto Shizuoka College of Pharmacy Japan
Saccharomyces cerevisiae ATCC 44668 - 44670 genetic	G. Simchen The Hebrew University Israel
Torulopsis pintolopesii ATCC 44696 causative agent of dermatophytic infection	I. Kunstyr Hannover Medical School Germany
Saccharomyces cerevisiae ATCC 44708 - 44724 genetic	R. Wickner NIH Bethesda, MD
Saccharomyces cerevisiae ATCC 44732 rapid ethanol fermentation	C.P. Kurtzman NRRL Peoria, IL
Saccharomyces cerevisiae ATCC 44769 - 44774 hosts certified by NIH for recombinant DNA research	D. Bolstein MIT Cambridge, MA

Organism / Strain / Subject	Depositor
Saccharomyces cerevisiae ATCC 44779 - 44784 genetic	A.J.S. Klar Cold Spring Harbor Laboratory Cold Spring Harbor, NY
Saccharomyces cerevisiae ATCC 44799 - 44800 genetic	N.M.S. Reddy BARC India
Candida paludigena ATCC 44801 taxonomy	W.I. Golubev USSR Academy of Science USSR
Pichia inositovora ATCC 44802 taxonomy	"
Saccharomyces cerevisiae ATCC 44804 - 44805 mutation in ornithine	C. White-Tabor NIH Bethesda, MD
Candida albicans ATCC 44806 - 44808 human pathogens	T.G. Mitchell Duke University Durham, NC
Saccharomyces cerevisiae ATCC 44809 - 44810 fatty acid requiring mutants	R. Roggenkamp University of Erlangen W. Germany
Saccharomyces cerevisiae ATCC 44820 - 44821 extrachromosomal rho$^-$ mutants	K. Karhonen Finnish For. Res. Inst. Finland
Saccharomyces cerevisiae ATCC 44822 - 44826 genetic	G. Simchen The Hebrew University Israel
Saccharomyces cerevisiae ATCC 44827 - 44828 genetic	C. Mclaughlin University of California Irvine, CA
Candida albicans ATCC 44829 - 44831 adenine-requiring auxotrophs	M. Pesti Hungarian Seed Trade Association Hungary
Lipomyces kononenkoae ATCC 44833 - 44838 high production of α-amylase	N. van Uden OEIRAS Portugal
Saccharomyces cerevisiae ATCC 44856 genetic	G. Simchen The Hebrew University Israel

Saccharomyces cerevisiae ATCC 44857 genetic	J.R. Johnston University of Strathclyde Scotland
Candida albicans ATCC 44858 - 44859 animal pathogens	J. van Cutsen Janssen Pharmaceuticals Belgium
Saccaromycopsis fibuligera ATCC 44872 soil isolate	T.M. Hammill State University of New York Oswego, NY
Saccharomyces cerevisiae ATCC 44876 - 44877 genetic	G.B. Koklhaw Purdue University W. Lafayette, IN
Saccharomyces cerevisiae ATCC 44878 - 44885 killer strains	K. Ouchi Research Institute of Brewing Japan
Saccharomyces cerevisiae ATCC 44941 - 44942 mating-types	J.F. Lemontt Oak Ridge Nat'l Lab Oak Ridge, TN
Saccharomyces cerevisiae ATCC 44953 produces benzo (a) pyrene hydroxylase	B. Kirsop NCYC England
Hansenula polymorpha ATCC 44954 - 44955 production of tryptophan	A.L. Demain MIT Cambridge, MA
Candida ethanolica ATCC 44956 new species	Czechoslovakia Culture Collection Bratislava Czechoslovakia
Saccharomyces carlsbergensis ATCC 44966 serological tests	N. Nishikawa Sapporo Breweries Ltd. Japan
Saccharomyces cerevisiae ATCC 44967 serological tests	"
Saccharomyces uvarum ATCC 44968 serological tests	"
Saccharomyces manchuicus ATCC 44969 serological tests	"

Culture	Depositor
Saccharomyces batatae ATCC 44970 serological tests	"
Saccharomyces pastorianus ATCC 44971 serological tests	"
Saccharomyces exiguus ATCC 44972 serological tests	N. Nishikawa Sapporo Breweries Ltd. Japan
Pichia rosa ATCC 44973 serological tests	"
Candida albicans ATCC 44984 - 44990 auxotrophic mutants	A. Sarachek Wichita State University Wichita, KS
Saccharomyces cerevisiae ATCC 46003 - 46009 used in recombination studies	A. Iskihawa Shizuoka University Japan
Saccharomycopsis lipolytica ATCC 46025 - 46028 genetic	D. Ogrydziak University of California Davis, CA
Saccharomyces cerevisiae ATCC 46029 - 46031 killer strains	A.L. Extrema University Granada Spain
Pichia guilliermondii ATCC 46036 produces alpha-galactosidase	J.J. Ellis NRRL Peoria, IL

II. Institute of Biochemistry and Physiology of Microorganisms, USSR Academy of Science, Pushchino, Moscow region I42292, USSR. Communicated by W.I. Golubev.

Recent publications:

Catalogue of cultures of microorganisms maintained in the institutes of the USSR. 1981. Publ. House "Nauka", Moscow, 246 pp.

Golubev, W.I., Manukian, A.R., Shkidchenko, A.N. 1981. Effect of growth conditions on capsule formation of yeast. Biol. J. of Armenia, 34, N 3: 259-264.

Golubev, W.I., V.M. Blagodatskaya, S.O. Suetin, R.S. Trotsenko. 1981. Pichia inositovora and Candida paludigena, two new species of yeasts isolated from peat. Int. J. Syst. Bacteriol., 31: 91-96.

III. Department of Food Science and Technology, University of California, Davis, California 95616. Communicated by H.J. Phaff.

Below follows the abstract of the Ph.D. dissertation by Donald L. Holzschu (1981). The work was done in the laboratories of H.J. Phaff and Dennis Hedgecock.

Molecular Taxonomy and Evolutionary Relationships Among Cactophilic Yeasts.

Summary

The taxonomic affinities and evolutionary relationships of cactophilic yeasts (those that have adapted to an existence in necrotic tissues of various cactus species) including Pichia heedii, the varieties of P. amethionina, the varieties of P. opuntiae, and P. cactophila have been studied. Traditionally, schemes to classify yeasts as well as other microorganisms on the basis of genetic relatedness have proven difficult because of a general lack of information pertaining to the speciation process. In addition, a fossil record is not available for analysis of the morphology of ancestral cells. Macromolecular analyses of the closely related but distinct cactophilic species of the yeast genus Pichia has corroborated earlier studies suggesting the evolution of these yeasts.

Over 70 strains comprising the cactophilic yeast species have been compared by DNA-DNA homology and an additional 340 strains have been analyzed for allozyme variation utilizing 14 enzyme systems. The two different metabolic types of P. heedii are very similar, i.e., they display greater than 95% DNA homology and a relatively small genetic distance value, 0.675. Pichia amethionina var. amethionina and P. amethionina var. pachycereana show a greater degree of divergence by nucleic acid analysis, i.e., approximately 65% DNA homology but greater similarity in allozyme pattern, genetic distance = 0.330. Results show that these varieties are not interfertile and therefore represent separate species. Also, it is apparent that genetic distance as measured by variation in structural proteins may not be a sensitive marker for determining overall genetic changes in organisms. DNA homology among P. opuntiae var. opuntiae and P. opuntiae var. thermotolerans is approximately 28% while the genetic distance value is 1.270. These yeasts are not interfertile and species status for P. opuntiae var. thermotolerans seems appropriate. The data presented above are discussed with regard to the host plant distribution of the yeasts, host plant chemistry, and the probable vectors of yeast dispersal. The combination of these diverse data directly supports the central dogma of macromolecular studies of microorganism evolution, namely, that macromolecular change reflects evolution and thus lineage.

Pichia cactophila strains are very similar, i.e., their genomes show greater than 85% DNA homology. However, genetic distances calculated from allozyme patterns can be large, i.e., generally less than but occasionally greater than 1.000. Evidence is presented that suggests that the type strain of P. cactophila has some DNA sequences not present in other strains of P. cactophila. This phenomenon has not been observed previously in DNA homology studies and the nature of the DNA in question

is unknown at present. The population structure of P. cactophila suggests that host race formation may be present among strains associated with different cacti. A group of strains very similar to but distinct from P. cactophila, [by a low (35%) degree of DNA homology and distinct allozyme pattern], has been recognized and will be described as a separate species, P. sp. PC.

The population structure of these cactophilic yeasts has been utilized to infer modes of speciation based on current models of speciation. The data suggest that the speciation process in at least some cases has been by rapid processes, whereas in others a slow gradual change in genetic material is resulting in genetic isolation. Also, from these data it appears that P. heedii, P. cactophila, and P. sp. PC entered the ecological niche provided by the columnar cacti independently from P. amethionina and P. opuntiae.

* * *

Dr. Eveline Gueho will return to France on February 1, 1982 after spending more than one year in our laboratory. A summary of her work will be given in the Spring issue of 1982. Drs. Leda Mendonca-Haglar and Allan Haglar are currently spending their sabbatical leave in our laboratory. They expect to return to Rio de Janeiro, Brasil in March, 1982. A summary of their work now underway will appear in the Spring issue of the Yeast Newsletter.

IV. Department of Plant Sciences, University of Western Ontario, London, Ontario, N6A 5B7, Canada. Communicated by A.W. Day.

Fimbriae in yeast-like fungi

We summarize here studies by A.W. Day and R.B. Gardiner on fimbriae in the Ustilaginales and more recently in ascomycetous yeasts. Surface protein fibrils similar to bacterial fimbriae or pili have been demonstrated in many yeasts and yeast-like species (4-9). The fimbriae initially described in the smut fungus, Ustilago violacea, are very long (over 20 µm), thin (6.5 nm), and flexuous. Over 20 other species of Ustilago and over 30 heterobasidiomycetous yeasts classified in the Ustilaginales (Rhodotorula, Leucosporidium, Rhodosporidium, Filobasidium, Bullera, Sporobolomyces, Sporidiobolus and Sterigmatomyces) have been shown to have this same type of fimbriae (5, 6). The protein involved has been isolated from a number of these species and appears to consist of two species each with M.W of 74,000 (2).

Antisera prepared against the pure fimbriae of U. violacea and Rhodotorula rubra agglutinate cells carrying these fimbriae. Tests of these antisera on about 50 Ustilaginales subdivided these species into five main categories (6).

Type I - afimbriate - no agglutination

Type 2, fimbriate - no agglutination

Type 3, fimbriate, agglutinated only by U. violacea antifimbrial antibodies

Type 4, fimbriate, agglutinated only by R. rubra antifimbrial antibodies

Type 5, fimbriate, agglutinated by both antibodies

Species of Ustilago showed a type 3 or type 5 response while the heterobasidiomycete species varied from Type I to Type 5, a result which may indicate the presence of a family of related surface proteins in this group (6).

Much shorter fimbriae have been noted on several ascomycetous yeasts including Saccharomyces cerevisiae and a correlation with flocculation was observed in brewer's strains (4, 8, 9). Recent observations (6) of several ascomycetous yeasts indicate that 1) nearly all examined species (in the genera Arthroascus, Saccharomyces, Kluyveromyces, Debaryomyces, Pichia, Hansenula, Nadsonia, Hanseniaspora and Metschnikowia, as well as related members of the deuteromycete, Candida) have a short fringe of fimbriae 0.5-1.0μm long, visible by shadowing water washed cells, but made clearer by washes with acetone; 2) many species including S. cerevisiae are agglutinated by crude or pure preparations of the 2 antisera derived from Ustilago and Rhodotorula (reactions of types II to V were observed); 3) as in heterobasidiomycetous species when visible fimbriation is temperature dependent, ability to be agglutinated by antisera shows a similar temperature dependency.

Fimbriae are therefore widespread in ascomycetous and basidiomycetous yeasts and a family of related proteins appears to be involved. This conclusion is supported by studies of fimbrial-defective mutants in Ustilago and Candida. Most of the mutants, selected for non-agglutination after antibody treatment, are temperature sensitive and show visible fimbriae and agglutinability only at high or low temperatures. A few mutants which do not agglutinate at any temperature have been found in these two species - as expected they lack visible fimbriae (Gardiner, MS in preparation).

There is no evidence so far that the Ustilago fimbriae contain any carbohydrates, but the ascomycete type may correspond to the manno-proteins known to predominate in the outer layers of yeast walls. Surface molecules such as fimbriae are likely to be multifunctional and roles for fungal fimbriae have been proposed for conjugation (1, 3, 7), pathogenicity (6), adhesion (4), flocculation (4, 9), and in the transport and functioning of large extracellular molecules (4).

References

1. Day, A.W. 1976 Nature 262:583-584.
2. Day, A.W. and J.E. Cummins 1981 In Sexual Interactions in Eukaryotic Microbes Academic Press pp. 379-402.
3. Day, A.W. and N.H. Poon 1975 Can. J. Microbiol. 21:547-557.
4. Day, A.W., N.H. Poon and G.G. Stewart 1975 Can J. Microbiol. 21:558-564.

5. Gardiner, R.B., M. Canton, and A.W. Day 1981 Bot. Gaz. 142:147-150.
6. Gardiner, R.B., K. Podgorski and A.W. Day 1982 manuscript in preparation.
7. N.H. Poon and A.W. Day 1973 Nature 250:648-649.
8. N.H. Poon and A.W. Day 1975 Can. J. Microbiol. 21:537-546.
9. N.H. Poon, B. Miki, V.L. Seligy and A.P. James 1978 Microscopical Society of Canada Vol. V pp. 60-61.

V. The University of Sydney, Department of Biochemistry, Sydney, N.S.W. 2006, Australia. Communicated by Audrey M. Bersten.

A short paper from this laboratory was recently published.

N.H. Packer and Audrey M. Bersten. Lipids and the morphogenesis of *Trigonopsis variabilis*. FEMS Letters 12:135-138 (1981).

Earlier investigations of the lipids of triangular cells of *Trigonopsis variabilis* induced by growth in methionine-medium, specified a role for phosphatidylcholine in determining the morphogenesis of the cell. Differences in the sterol composition of the triangular and ellipsoidal cells have been observed. Triangle-shaped cells can be induced by only Tween 80, among the Tween series of detergents. This is due to the sorbitan mono-oleate moiety of the Tween, suggesting a role for oleate in the shape determination.

When *Trig. variabilis* was grown in continuous culture in methionine-medium a specific growth rate and temperature were necessary for the development of triangular cells. Growth rate and temperature modify the lipid composition of a variety of yeasts. In order to differentiate between changes in lipid content resulting from an alteration to the growth medium and changes actually producing the different morphology, the lipids of the same shape of cell induced by different environmental factors are compared.

Homoazasterol is an inhibitor of sterol synthesis in *Saccharomyces cerevisiae*. The effect of this fungicide on triangular cell development is also presented.

VI. Department of Plant Sciences, University of Western Ontario, London, Ontario, N6A 5B7, Canada. Communicated by M.A. Lachance.

The following are abstracts of recent papers from this laboratory.

Deborah Gayle Sidenberg and Marc-André Lachance. Electrophoretic patterns of exo-β-glucanase in *Kluyveromyces* species: evidence for multiple molecular forms. Experimental Mycology (in press).

Abstract

Exo-β-glucanases (EC 3.2.1.58) of the type strains of the yeast genus *Kluyveromyces* were examined by polyacrylamide electrophoresis. Five

species were found to possess electrophoretically distinct multiple molecular forms. K. marxianus had two bands of exo-β-glucanase which showed identical activity ratios when assayed with laminaran and with p-nitrophenyl-β-D-glucopyranoside as substrates. Their charges appeared similar, but their molecular weights differed by 5 kilodaltons. A single band of activity was detected in K. bulgaricus, and it appeared essentially identical to the fast band of K. marxianus. Three molecular forms were observed in K. drosophilarum. A histochemical staining method for glucohydrolases is described.

* * *

Marc-André Lachance and William T. Starmer[1]. Evolutionary Significance of Physiological Relationships Among Yeast Communities Associated with Trees. Canad. J. Bot. (in press).

[1]Department of Biology, Syracuse University, Syracuse, NY 13210

Abstract

The physiological attributes of yeast communities associated with trees from 1 Gymnosperm and 9 Angiosperm families were examined in comparison with contemporary classifications of the tree taxa. While the relationships between yeasts found in 5 coniferous genera (Pinaceae) did not show much congruence with chemotaxonomic or immunological classifications, a physiological classification of yeast communities from 19 genera of Angiosperms showed some compatibility with their respective assignments to families. Clustering of trees as yeast habitats at the family and higher levels indicated that the physiological profiles of yeast communities are to some degree correlated with the taxonomic position of the tree taxa as viewed by some authors. The yeast communities associated with certain tree families appeared nutritionally specialized as compared to others. This evolutionary specialization most likely operates by selection of physiologically limited yeasts, rather than by reduction of the number of yeast species in each community.

* * *

M.A. Lachance. 1981. Hanseniaspora nodinigri, a new yeast species found in black knots (Dibotryon morbosum) of Prunus virginiana. Canad. J. Microbiol. 27:651-653.

Abstract

The new yeast species Hanseniaspora nodinigri is described to accommodate members of the genus Hanseniaspora that are unable to assimilate glucono-δ-lactone and isolated from stromatal tissue of black knots (Dibotryon morbosum) of chokecherry, Prunus virginiana. The newly described taxon shows much resemblance, by other criteria, to H. vineae van der Walt et Tscheuschner and H. osmophila (Niehaus) Phaff, Miller et Shifrine.

VII. National Research Council Canada, Division of Biological Sciences, Ottawa, Canada K1A OR6. Communicated by Byron F. Johnson.

Below follow two abstracts from our laboratory.

Byron F. Johnson, Gode B. Calleja, Bong Y. Yoo, Michael Zuker and Ian J. McDonald. Cell Division: Key to Cellular Morphogenesis in the Fission Yeast, *Schizosaccharomyces*. International Review of Cytology, 75: (in press), 1982.

Abstract

Cell division of the fission yeast is reviewed cytologically, and then as a cellular process having morphogenetic and physiological consequences notable in succeeding cell cycles. Anomalies of cell division are considered to be breakdowns of controls, hence are compared in order to illustrate some features of cell cycle regulation related to cell division.

* * *

Temporal asymmetry of sex interconversion in a strain of the homothallic fission yeast, *Schizosaccharomyces pombe*. Current Microbiology 6: 227-230, 1981.

Abstract

In the homothallic P/d interconversion system of the fission yeast, *Schizosaccharomyces pombe*, P → d is apparently twice as frequent as d → P. This is interpreted to mean that P → d occurs before DNa replication whereas d → P occurs after. But the probabilities of their occurrence within a cell cycle are about the same (1 in 2^7 cell divisions).

VIII. Department of Microbiology, Faculté de Pharmacie, Avenue Charles Flahault, 34060 Montpellier Cedex, France. Communicated by J.M. Bastide.

Below follow titles of recent papers and posters from this laboratory:

1. M. Bastide, S. Jouvert, J.M. Bastide. Action des antifongiques sur la paroi et la membrane cytoplasmique de *C. albicans* révelée par microscopie électronique à balayage. I. - Amphotericine B et Nystatine. Bull. Soc. Mycol. Med., 1981, X, 113-118.

Summary

Nystatin (50 U.I. and 500 U.I. ml^{-1}) or amphotericin B (50 µg and 500 µg ml^{-1}) acting on intact yeasts or sphaeroplasts of *Candida albicans* challenged important lesions on the cytoplasmic membrane, but not on the cell-wall, as revealed by scanning electron microscopy.

* * *

2. M. Bastide, S. Jouvert, J.M. Bastide. Cell wall and cytoplasmic membrane attack level by polyene antibiotics and imidazoles: study on Candida albicans by S.E.M. VIIth. Internat. Specialized Symp. on Yeasts, Valencia, September 23, 1981. Spain.

Abstract

Young yeasts or sphaeroplasts (3 hours old) were treated by antifungal solutions during 2 h., 28°C:

Polyenes: amphotericine B, 500 $\mu g.ml^{-1}$, 50 $\mu g.ml^{-1}$
nystatin 500 U.I. ml^{-1}, 50 U.I. ml^{-1}

Imidazoles: clotrimazole, econazole, miconazole, isoconazole, ketoconazole: 500 $\mu g.ml^{-1}$, 50 $\mu g.ml^{-1}$

The action of these antibiotics was evaluated on the cell wall (yeasts) and on the cytoplasmic membrane (sphaeroplasts). When going through the cell-wall the localization of these substances was studied by examining released sphaeroplasts after action on the whole yeasts. These conditions (high concentrations, short contact) allowed us to observe the early attack of the antifungal agents. The observations were performed with scanning electron microscope (JSM. 35).

The polyenes showed a localization on cytoplasmic membrane leading to a disruption of the sphaeroplasts. By contrast, the cell wall appeared to be intact.

The imidazoles acted very weakly on the cytoplasmic membrane, but modified the cell wall and the releasing of sphaeroplasts. We could separate the imidazoles in three groups, according to some differences of action: one including ketoconazole, one another including clotrimazole, and the third gathering econazole, isoconazole, miconazole.

All these results may demonstrate an activity-structure relation.

* * *

3. J.M. Bastide, S. Jouvert, J.C. Lebecq and M. Bastide. Surface ultrastructure variation of C. albicans sphaeroplasts. VIIth Internat. Specialized Symp. on Yeasts, Valencia, September 23, 1981, Spain.

IX. Yamaguchi University, Biological Institute, Faculty of Science, Yamaguchi 753, Japan. Communicated by Nobundo Sando.

The following papers have been published or will appear very soon.

Nobundo Sando, Tomoko Oguchi, Misuzu Nagano and Masako Osumi. Morphological changes in ascospores of Saccharomyces cerevisiae during aerobic and anaerobic germination. Journal of General and Applied Microbiol. 26:403-412 (1980).

Abstract

The stainability of ascospores and vegetative cells of Saccharomyces cerevisiae to acid-fast staining, using hot Ziehl's carbolic fuchsin solution, 5% sulfuric acid, and diluted Loffler's methylene blue, was examined. Resting spores and growing haploid cells retained much fuchsin dye in the cells. Only mature spores of the diploid strain resisted methylene blue staining. The stainability of Mycobacterium phlei IFO 3158 also was examined.

The kinetics of germination was examined. The loss of stainability with acid-fuchsin and of resistance to methylene blue was used as a criterion of germination. The ascospores germinated anaerobically as well as aerobically. Especially in the early stage of germination, there was found no difference in the germination rates under either conditions. Ultrastructure of germinating ascospores cultured in aerobic and anaerobic conditions was examined by ultrathin sectioning and electron microscopy. At the first stage of germination, the ascospores swelled in aerobic as well as anaerobic cultures. The outer spore coat and the outer zone of the inner spore wall disappeared during the germination process, the inner zone of the spore wall then giving rise to a germinated spore cell wall (extruded germ tube wall). The vacuole became granular. The mitochondria showed no change in shape and number in aerobic cultures, but seemed to swell and disintegrate in the later stages of anaerobic germination.

* * *

Nobundo Sando, Isamu Miyakawa, Sohryu Nishibayashi and Tsuneyoshi Kuroiwa. Arrangement of mitochondrial nucleoids in life cycle of Saccharomyces cerevisiae. Journal of General and Applied Microbiology 27 (6) (1981). Published in December.

Abstract

In synchronously growing or sporulating cells of Saccharomyces cerevisiae, nuclei and mitochondrial DNA (mtDNA) were stained with fluorescent dye, 4', 6-diamidino-2-phenylindole (DAPI) with a buffer-NS, containing sucrose, EDTA, $MgCl_2$, $ZnSO_4$, $CaCl_2$, PMSF and mercaptoethanol. They were examined under a modified Olympus epi-fluorescence microscope BH-2. Many mtDNAs were observed being separated from each other in cells at stationary phase. In meiosis I or early budding, the mtDNAs aggregated and formed "string-of-beads" appearance, as described by D.H. Williamson and D.J. Fennell (1975) and B.J. Stevens (1978). Especially in meiosis II, the arrays of mtDNAs formed some replicating figures attached at each end to a haploid nucleus. The "strings" of mtDNA as a whole increased the fluorescence and transiently formed uniformly wide bands. Most part of the "string-of-beads" of mtDNAs was enclosed into a spore, surrounding the spore nucleus, and the other part remained in the epiplasma to be degraded. In budding cells, on the other hand, mtDNAs were integrated in a single string and transferred into a new bud cell. The thread fragmented again into separate nucleoids in the daughter and mother cells.

X. Institut National Agronomique, Chaire de Génétique 16, Rue Claude-Bernard, 75231, Paris-5, France. Communicated by H. Heslot.

Below follow abstracts of recent works from our Institute.

J.M. Beckerich, M. Lambert and H. Heslot. Mutations affecting simultaneously the lysine and polyphosphate pools in the yeast Saccharomycopsis lipolytica. Biochem. Biophys. Res. Comm. 100:1292-1298. 1981.

A specific staining procedure was devised to screen for polyphosphate minus mutants (ply) in the yeast Saccharomycopsis lipolytica. These mutants were specifically altered in the polyphosphates classes of high molecular weight. Moreover, the mutants showed a decreased ability to retain lysine. However, there was no stoichiometric relationship between lysine and polyphosphate pools. The storage of lysine appeared in many cases fully uncoupled from the pool of polyphosphates, excluding that polyphosphates play a role as cationic receptors. On the other hand, the lysine and polyphosphate pools were simultaneously depleted in the mutants, suggesting some kind of relationship between these pools.

* * *

C. Gaillardin, P. Fournier, L. de Louvencourt, H. Heslot and C. Gerbaud (Laboratoire Génétique et Biologie Moleculaire, Fac. Sciences, Bat. 400, 91405 Orsay Cedex, France). Behavior of S. cerevisiae replicators in Schizosaccharomyces pombe (manuscript in preparation).

Transformation of the yeast Schizosaccharomyces pombe using a whole 2 µm plasmid and a $LEU2^+$ gene of Saccharomyces cerevisiae has been reported recently (Beach & Nurse, 1981, Nature, 290, 140). We investigated the efficiency of various yeast replicators in this respect, as well as the fate of the transforming plasmids.

A 2 µm plasmid of S. cerevisiae associated with a marker gene ($URA3^+$) of the same yeast was used for transformation of Schizosaccharomyces pombe, strain ura4.294h$^-$. Prototrophic transformants were obtained at a frequency of 10^{-5} transformants per µg of DNA per viable protoplasts, but up to 80% clones were abortive. The surviving clones were unstable, and the degree of stability varied depending on the site used for cloning the 2 µm in pBR 322-$URA3^+$ and on the form (A or B) of the 2 µm plasmid. The structure of the plasmids present in the transformants has been investigated. It turned out that all types of plasmids underwent gross rearrangements in S. pombe. Moreover, transformation was successfully achieved with plasmids containing only part of the 2 µ DNA, bearing or not the sequence required as an origin of replication in S. cerevisiae. We are currently investigating the fate of the transforming DNA in these cases.

Chromosomal replicators of S. cerevisiae-like ars1 or ars2 were also found to be effective for transformation of S. pombe, resulting in a high number of unstable transformants. The plasmids recovered in E. coli were unmodified.

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P. Fournier, C. Gaillardin, L. de Louvencourt, H. Heslot, and B. Lang and F. Kaudewitz (Genetisches Institut des Universität München, Maria Ward Str 1a, 8000 München 19, GFR). Extrachromosomal rDNA from S. pombe: Cloning and use for transformation (manuscript in preparation).

We have isolated a naturally occurring plasmid of about 3 μm length, which was found associated with crude mitochondrial fractions of S. pombe. It has been cloned in pBR 322 and a restriction map established; its size is about 10.8 kb and possesses unique sites for the enzymes Hind III, Bgl II, Hpa I, Sal I. Associated with the URA3 gene from S. cerevisiae, it is able to transform a S. pombe ura4 mutant to URA^+ at a rather low frequency. It is maintained as an extrachromosomal element as we could recover it by back transformation in E. coli. The plasmid was not rearranged except in one case where a gross deletion had occurred. Hybridization of this plasmid DNA with "ribosomal" DNA from S. cerevisiae revealed sequence homologies in DNA segments coding for the ribosomal RNA subunits of S. cerevisiae.

XI. Division of Infectious Diseases, UCLA School of Medicine, Department of Medicine, E-5, Torrance, California 90509. Communicated by Marjorie Crandall.

Below follow abstracts of two recently submitted papers.

Marjorie Crandall and D. Vincent Waterhous. Virulence of a Methionine Auxotroph of Candida albicans. Infection and Immunity (submitted).

Abstract

Virulence of an isogenic set of four C. albicans strains was determined in mice. The four isogenic strains were: a clinical isolate naturally heterozygous for methionine (+/met), a homozygous auxotrophic (met/met) segregant, a prototrophic (+/+) segregant and a revertant (+/met). The met/met and +/+ segregants are reciprocal recombinants derived from a sectored colony resulting from UV-induced mitotic recombination. No difference in the lethal time for 50% of mice (LT_{50}) was observed for these four strains indicating that a requirement for methionine has no effect on virulence. This conclusion may be drawn because a comparison of strains within an isogenic set allows for possible variations in the genetic background of the mutant to be controlled and analyzed. The significance of the diploid/disomic state of C. albicans and spontaneous mitotic segregation is discussed with regard to mechanisms of pathogenesis by this opportunistic yeast.

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Marjorie Crandall. UV-induced Mitotic Co-segregation of Genetic Markers in Candia albicans. Evidence for Linkage. Current Genetics (submitted).

Summary

UV-induced mitotic recombination is a valuable tool for parasexual genetic studies of medically important yeasts. Low dose UV-irradiation of the Hoffmann-La Roche type strain of C. albicans (#6631) yielded a limited spectrum of mutants at a high frequency. Canavanine (can) sensitive segregants were induced at a frequency of 7.6 x 10^{-3}. Double mutants that were both can-sensitive and methionine (met) auxotrophs occurred at a frequency of 7.4 x 10^{-3}. The expected single met segregant class was missing. UV-induced can and met can segregants occurred in sectored colonies. Analysis of sectors led to the conclusion that strain 6631 is naturally heterozygous for met and can which are linked in the cis configuration. The proposed gene order is: centromere - met - can.

XII. Pabst Brewing Company, Brewing Research & Development*, Milwaukee, Wisconsin 53201 and the Department of Genetics, Ohio State University, Columbus, Ohio 43210. Communicated by Michael F. Waxman.

Michael F. Waxman* and C. William Birky, Jr. Partial Pedigree Analysis of the Segregation of Yeast Mitochondrial Genes During Vegetative Reproduction. Submitted to Current Genetics for publication.

Abstract

A three-factor cross of Saccharomyces cerevisiae involving the capl, eryl, and olil loci was done, with partial pedigree analysis of 117 zygotes. First, second and third buds were removed and the genotypes of their diploid progeny determined, along with those of the residual zygote mother cells. Results were analyzed in terms of frequencies of individual alleles and of recombinant genotypes in the dividing cells. There is a gradual increase in frequency of homoplasmic cells and in gene frequency variance during these three generations, as would result from stochastic partitioning of mtDNA molecules between mother and bud, probably coupled with random drift of gene frequencies in interphase cells. These phenomena are more pronounced for buds than for mothers, suggesting buds receive a smaller sample of molecules. End buds are more likely to be homoplasmic and have a lower frequency of recombinant genotypes than do central buds; an end bud is particularly enriched in alleles contributed by the parent which formed that end of the zygote. These results suggest that mixing of parental genotypes occurs first in the center of the zygote. Zygotes with first central buds produce clones with a higher recombination frequency than do those with first end buds. If segregation were strictly random, the number of segregating units would have to be much smaller than the number of mtDNA molecules in the zygote. On the other hand, there is no evidence for a region of the molecule ("attachment point") which segregates deterministically.

XIII. The University of Tokyo, Department of Agricultural Chemistry, Bunkyo-Ku, Tokyo 113, Japan. Communicated by Takemitsu Mizunaga.

Below follows the summary of a recent paper from our laboratory.

Takemitsu Mizunaga and Toshihiro Noguchi. The Role of Core-Oligosaccharide in Formation of an Active Acid Phosphatase and its Secretion by Yeast Protoplasts. Journal of Biochemistry, Vol. 91, No. 1 (January) (1982) (in press).

Summary

Tunicamycin (TM), an antibiotic that blocks glycosylation of glycoproteins by inhibiting the formation of dolichyl N-acetylglucosaminyl pyrophosphate was used to study the expression of active repressible acid phosphatase (r-APase) (EC 3.1.3.2) by yeast protoplasts. Secretion of active r-APase was completely inhibited by TM; however, neither accumulation of r-APase activity inside protoplasts nor inhibition of protein synthesis were observed on TM-treatment. The results led us to postulate that an enzymatically inactive form of nonglycosylated r-APase is accumulated in the membrane fraction on TM-treatment. Protoplasts of various r-APase mutants were radiolabelled with (S^{35}) methionine in the presence or absence of TM, then membrane fractions were analyzed by sodium dodecyl sulfate-polyacrylamide-gel electrophoresis and an autoradiogram was prepared. In r-APase-producing protoplasts of the wild type strain, the membrane (100,000 x g sedimentable) fraction of TM-treated protoplasts contained, in addition to usual membrane proteins, three proteins with molecular masses of 59K, 57 K and 55 K daltons. The three proteins are also formed in TM-treated protoplasts of the pho80 mutant constitutively forming r-APase in the presence of Pi. However, TM did not induce formation of these three proteins in the protoplasts of r-APase negative mutants (pho2, pho4 and pho81), or in the protoplasts of temperature sensitive mutants ($pho2^{ts}$ and $pho4^{ts}$) at a nonpermissive temperature (35°), or in TM-treated protoplasts of the wild type strain repressed by Pi. The three proteins were located in the membrane fraction and not in the cytosol or outside of the protoplasts. The purified r-APase preparation deglycosylated by hydrogen fluoride (HF) treatment at 0°C for 1 hr or by endo-β-N-acetylglucosaminidase H (Endo H) treatment, yielded three proteins with molecular masses of 62K, 60K, 58K or 60K, 58K, 56K daltons, respectively. We considered the purified r-APase preparation to be composed of three different molecules, which yielded three deglycosylated proteins. The difference of about 2000 daltons in apparent molecular mass between the deglycosylated proteins produced by HF and Endo H treatments is probably due to N-acetylglucosamine residues attached to asparagine residues in the r-APase proteins.

Our results indicate that the 59K, 57K and 55K proteins produced by TM-treatment are nonglycosylated forms of r-APase and that addition of core-oligosaccharide is required for formation of active r-APase and its secretion by yeast protoplasts.

XIV. Albert-Ludwigs Universität, Biochemisches Institut, Hermann-Herder-Str. 7, D-7800 Freiburg I. Br., West Germany. Communicated by Dieter H. Wolf.

Below follows the summary of our recent work concerning the isolation and analysis of proteinase A mutants of *S. cerevisiae*.

Bernd Mechler and Dieter H. Wolf. Analysis of Proteinase A Function in Yeast. Eur. J. Biochem. in press.

Summary

Yeast mutants lacking proteinase A were isolated. One of these mutants (HbI) is characterized in detail. The mutation called pral segregates 2:2 in meiotic tetrads indicating a single gene mutation. No proteinase A - antibody cross reacting material can be detected. Diploids heterozygous for pral show gene dosage. Thus, it appears that PRA1 might be the structural gene for proteinase A.

Results obtained with this mutant show that proteinase A is not a vital component of the vegetative cell cycle. The mutant exhibits normal mitotic growth under rich and poor growth conditions and shows normal mating. Enzymes subject to carbon catabolite inactivation and inactivation of NADP-dependent glutamate dehydrogenase, processes which were proposed to be of proteolytic nature, are not affected by the absence of proteinase A. However, protein degradation under sporulation conditions is about 30% reduced in proteinase A mutant cells. The differentiation process of sporulation is also disturbed leading to a 40% reduced sporulation frequency in mutant cells.

XV. Osaka University, Department of Fermentation Technology, Faculty of Engineering, Osaka University, Yamada Kami Suita-Shi, Osaka, Japan. Communicated by Yasuji Oshima.

Below follow abstracts of two recent papers from our Department.

Akio Toh-E, S. Inouye, and Y. Oshima (1981). Structure and Function of the PHO82-pho4 Locus Controlling the Synthesis of Repressible Acid Phosphatase of Saccharomyces cerevisiae. J. Bacteriol. 145:221-232.

pho4 mutants of Saccharomyces cerevisiae, although rare among phosphatase-negative mutants isolated from wild-type strains, were isolated efficiently from pho80, pho85, or pho80 pho85 strains. The distribution of these pho4 mutants over the pho4 locus was determined by analyzing random spores of two- and three-factor crosses. The pho4-4 mutation confers temperature-sensitive synthesis of repressible acid phosphatase. An intragenic suppressor for the pho4-12 allele results in the temperature-sensitive synthesis of repressible acid phosphatase. Recombination between these sites occurs at 1.0 to 3.0%, the highest for any pair of sites within the pho4 locus. All these results strongly indicate that the information of the pho4 locus is translated into a protein. The PHO82 site was mapped inside the pho4 locus by random spore analysis. The order met10-pho4-1PHO82-1-pho4-9 on the right arm of chromosome VI was confirmed by tetrad analysis. Doubly heterozygous diploids, pho3 PHO82^{c} PHO4^{+}/pho3pho82^{+} pho4, produce variable amounts of repressible acid phosphatase under repressive conditions depending on the combination of PHO82^{c} and pho4 alleles. This phenomenon may reflect the constitutive production of the pho82^{+}-pho4 product in the repressed condition, which interferes with the function of the PHO82^{c}-PHO4^{+} product. The earlier model for the function of the PHO82-pho4 cluster, in which the PHO82 site acts as an operator of the pho4 gene, has been

revised to a model in which the PHO82 site codes for the part of the pho4 protein that has affinity for the regulatory protein encoded by the pho80 and pho85 genes.

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Kunihiro Matsumot, Akio Toh-E, and Yasuji Oshima 1981. Isolation and Characterization of Dominant Mutations Resistant to Carbon Catabolite Repression of Galactokinase Synthesis in Saccharomyces cerevisiae. Mol. Cell. Biol. 1:83-93.

Seven dominant mutations showing greatly enhanced resistance to the glucose repression of galactokinase synthesis have been isolated from GAL81 mutants, which have the constitutive phenotype but are still strongly repressible by glucose for the synthesis of the Leloir enzymes. These glucose-resistant mutants were due to semidominant mutations at either of two loci, GAL82 and GAL83. Both loci are unlinked to the GAL81-gal4, gal80, or gal7 gal10 gal1 locus or to each other. The GAL83 locus was mapped on chromosome V at a site between arg9 and cho1. The GAL82 and GAL83 mutations produced partial resistance of galactokinase to glucose repression only when one or both of these mutations were combined with a GAL81 or a gal80 mutation. The GAL82 and GAL83 mutations are probably specific for expression of the Leloir pathway and related enzymes, because they do not affect the synthesis of α-D-glucosidase, invertase, or isocitrate lyase.

XVI. Department of Botany, University of Baluchistan, Quetta, Pakistan. Communicated by S.R. Chughtai.

We have tested two of the major constituents of betel quid, betel leaf and betel nut, for their possible mutagenic and recombinogenic activity in diploid yeast. A paper about the mutagenicity testing of betel leaf has been submitted to Mutation Research. Below follows a summary of the paper that we intend to present at "All Pakistan Conference of Plant Scientists" at Karachi Pakistan, to be held in February, 1982.

S.R. Chughtai, A.S. Mohamand and B.A. Siddiqui. Evaluation of Possible Mutagenicity and Recombinogenicity of Betel Nut in Diploid Yeast.

Betel nut is a major constituent of betel quid. However, its role in the induction of oral cancers, associated with the habitual chewing of betel quid, has not been established experimentally. We have evaluated the betel nut for its possible mutagenicity and recombinogenicity in diploid yeast. The aqueous extract of betel nut was tested for the induction of mitotic gene conversion, crossing over and reverse mutation in diploid strains D4 and D7 of Saccharomyces cerevisiae. The tests were performed with stationary as well as with log-phase cells. In stationary-phase tests, the treatments were carried out at two different pH levels: 5.91 and 8.05. The results indicate that betel nut, without any further metabolic activation, was non-mutagenic and non-recombinogenic. If failed to increase significantly the frequency of convertants, mitotic recombinants and revertants in diploid yeast. Moreover, no significant cell killing or inhibition of cell division was observed. EMS, used as a positive control, exhibited mutagenic and recombinogenic activity.

XVII. College of Medicine and Dentistry of New Jersey, Rutgers Medical School, Department of Microbiology, University Heights, Piscataway, New Jersey 08854. Communicated by Michael J. Leibowitz.

Below follow abstracts of two manuscripts recently completed in our laboratory.

Localization of Genes on the Double-stranded RNA Killer Virus of Yeast. J. Douglas Welsh* and Michael J. Leibowitz. Proc. Natl. Acad. Sci. U.S.A., in press.

*Present address: Department of Microbiology, School of Medicine, State University of New York at Stony Brook, Stony Brook, New York 11794.

Abstract

The M dsRNA genome segment of the cytoplasmically-inherited killer virus of yeast codes for two polypeptides when denatured and translated *in vitro*: these have molecular weights of 32,000 and 19,000 daltons by $NaDodSO_4$-polyacrylamide gel electrophoresis. An internal 190 base-pair region of the dsRNA is selectively degraded by S1 nuclease treatment at 65°C, resulting in two dsRNA fragments which contain the termini of the original dsRNA. The larger fragment codes for the 32,000 dalton polypeptide while the smaller fragment codes for the 19,000 dalton polypeptide. Thus, the two gene products of M are encoded by distinct regions of this dsRNA.

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Separation and Sequence of the 3' Termini of M Double-stranded RNA from Killer Yeast. Dennis J. Thiele, Regina W. Wang, and Michael J. Leibowitz. Submitted for publication.

Abstract

Four subspecies of M double-stranded RNA from a killer strain of *Saccharomyces cerevisiae* were isolated. Each subspecies was susceptible to heat cleavage, presumably at an internal 190 base pair A,U-rich region, generating two discrete fragments corresponding to each side of the A,U-rich region. Enzymatic and chemical RNA sequence analysis defined the 3'-terminal 175 bases for the larger fragment (M-1) and 231 bases for the smaller fragment (M-2). All four subspecies of M have identical size and 3'-terminal sequences. Potential translation initiation codons are present on the corresponding 5' termini of both fragments, and a possible 18S ribosomal RNA binding site is also present on the 5' terminus of M-1. Stem and loop structures for the 5' and 3' termini of M-1 may function as recognition sites for replication, transcription, and translation.

XVIII. Centre National de La Recherche Scientifique, Laboratoire D'Enzymologie, 91190 Gif Sur Yvette (France). Communicated by J. Schwencke.

The following paper has been recently published.

M. Pilar Fernandez, Santiago Gascon and Jaime Schwencke. Some enzymatic properties of vacuolar alkaline phosphatase from yeast. Current Microbiology 6: 121-126 (1981).

After working for some time with yeast vacuoles, we are presently working in the field of proteolysis in yeast. We are mainly interested in the study of the proteolytic activities in yeast after UV-irradiation in collaboration with Dr. Ethel Moustacchi (Institut Curie, Section Biologie, Centre Universitaire d'Orsay).

We are also interested in the search of new protease activities. In collaboration with Dr. Maripaz Suarez and Miss Nieves Garcia from the group of Dr. Santiago Gascon (Universidad de Oviedo, Spain) we have recently found a new protease activity bound to an insoluble fraction of a crude yeast extract. The following is an abstract of a recent publication.

1. M.P. Suarez, J. Schwencke, N. Garcia and S. Gascon. A new X-prolyl-dipeptidyl aminopeptidase from yeast associated with a particulate fraction. FEBS Letters 131, 296-300, 1981.

A new proteolytic activity in Saccharomyces cerevisiae has been found partly associated with a 100,000 xg particulate fraction. We propose to denominate this activity X-prolyl-dipeptidyl aminopeptidase because it liberates p-nitroanilide from the synthetic dipeptides L-alanyl-L-proline-p-nitroanilide and glycyl-L-proline-p-nitroanilide. Neither prolyl-p-nitroanilide nor N-benzyloxycarbomyl-L-alynyl-L-proline-p-nitroanilide are substrates for the enzyme. The same levels of activity are present in the pleiotropic mutant S. cerevisiae 20B-12 having very low levels of proteinases A, B and carboxypeptidase Y, when compared to its wild-type strain X2180-1B.

Studies to further characterize this activity and to define its cellular localization are in progress. These studies will help to elucidate its physiological role which according to its particulate nature could be associated with membrane translocation or other membrane-associated proteolytic phenomena.

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The following is an abstract of a paper recently accepted for publication in Analytical Biochemistry.

2. J. Schwencke. Measurement of proteinase B activity in crude yeast extracts. A novel procedure of activation using pepsin. Analytical Biochemistry, 118 (1981) in press.

Proteinase B activity in crude yeast extract is difficult to detect unless the firmly bound peptide inhibitor is previously inactivated. Methods currently used to accomplish this inactivation suffer from serious limitations, particularly when applied to crude extracts obtained from yeast grown in complete media. These limitations are discussed on the basis of the kinetics of activation.

A novel procedure to unmask proteinase B activity, based on the pepsin treatment of crude extracts, is described. Specific activity of proteinase B, determined after pepsin A treatment, is always two to three times higher than that obtained by other methods. The method proposed is simple, reliable, and can be applied to crude extracts stored at -20°C for up to 1 week, repeatedly giving the same specific activity for proteinase B.

XIX. Instituto de Enzimologia Y Patologia, Molecular Del C.S.I.C., Facultad de Medicina de la Universidad Autonoma, Arzobispo Morcillo, S/N, Madrid 34, Spain. Communicated by C. Gancedo.

Below follows the summary of an article that will appear in J. Biol. Chem. February, 1982.

Mazon. M.J., Gancedo, J.M. and Gancedo, C. Inactivation of yeast fructose-1,6-bisphosphatase. In vivo phosphorylation of the enzyme.

Summary

Incorporation of ^{32}P into yeast fructose-1,6-bisphosphatase (E.C.3.1.3.11.) was observed after addition of glucose to a cell suspension incubated with ^{32}P orthophosphoric acid. The ^{32}P counts were coincident with the enzyme band when immunoprecipitates were subjected to sodium dodecyl sulfate disc gel electrophoresis. The incorporation of phosphate was associated with a decrease in enzyme activity. Approximately one mole of phosphate was incorporated per mole of enzyme. The phosphate is bound to the enzyme in a phosphoester linkage with a serine residue. Release of ^{32}P accompanying enzyme reactivation was observed both in vivo and in cell free extracts.

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Lagunas, R., A. Busturia, and C. Dominguez. Inactivation of Sugar Transport Systems in S. cerevisiae and its Relation to the Pasteur Effect. VIIth International Specialized Symposium on Yeast Surface, 23-27 September 1981, Valencia, Spain.

Pasteur effect (activation of sugar consumption by anaerobiosis) is irrelevant in growing S. cerevisiae. The phenomenon appears only after exhaustion of the nitrogen source in the medium (R. Lagunas. T.I.B.S. in press). The reasons for this different behaviour of growing and resting cells have been studied in various yeast strains and the results suggest the following conclusions:

Contribution of respiration to sugar catabolism is small in growing yeast. However, upon disappearence of a nitrogen source the ability to ferment sugars greatly decreases and respiration becomes preponderant. A shift to anaerobiosis of these cells would decrease the formation of ATP which tends to be compensated by an increase of the rate of sugar consumption.

The responsible mechanism for the decrease of fermentation in resting cells has also been studied. Two factors contribute to this decrease: a

direct effect of the lack of ammonium on phosphofructokinase and a rapid inactivation of the sugar transport systems due to their rapid turnover. The following approximate half-lives have been observed:

Glucose transport system, 7 hr.
Maltose transport system, 3 hr.
Galactose transport system, 3 hr.

These values contrast with the 70 hr. half-life of the pool of proteins in yeast and suggest a preferential degradation of the sugar carriers.

Addition of ammonium to resting yeast restores sugar transport activities as well as glycolytic flux in about two hours. Since the increase of total proteins is negligible during this period, it appears that yeast synthesizes the transport systems before resuming normal growth.

XX. Instituto de Enzimologia y Patologia Molecular. Facultad de Medicina. Universidad Autonoma de Madrid, Madrid-34, Spain. Communicated by Rosario Lagunas.

Is Saccharomyces cerevisiae a typical facultative anaerobe? Trends in Biochemical Sciences, (1981) 6, 201-203.

YEAST 12-81

Summary

Facultative anaerobes are usually defined as having three particular characteristics: (i) the ability to grow aerobically or anaerobically using oxygen or organic compounds as the final acceptors of electrons produced in catabolism; (ii) the preferential use of oxygen, if available, due to the greater energy yield obtained from the fuel molecules (36 mol of ATP/mol of hexose obtained in respiration v. 2 in fermentation) and; (iii) the smaller rate of breakdown of sugars (glucose is generally mentioned) in aerobiosis than in anaerobiosis as a consequence of the great difference in ATP yield. This is also stated as an inhibition of fermentation by respiration, the so called 'Pasteur effect'.

It is my purpose to draw attention to the fact that the yeast Saccharomyces cerevisiae, considered as a model facultative anaerobe, does not meet the above criteria. (i) Although S. cerevisiae can grow aerobically or anaerobically, anaerobic growth can only last for a few generations as the yeast requires molecular oxygen for the synthesis of ergosterol and unsaturated fatty acids, both components of its membranes; (ii) During growth on sugars (particularly on glucose) S. cerevisiae scarcely respires and fermentation is the main catabolic pathway even if oxygen is available. Actually, about 95% of the catabolized glucose, fructose, or maltose is fermented. Therefore, as far as these sugars are concerned, aerobiosis is energetically almost irrelevant. Even during growth on galactose, where yeast shows a greater respiratory capacity, the effect of oxygen on functions which would be affected by the amount of available ATP is almost negligible. This does not mean that aerobiosis is always energetically unimportant to yeast. Indeed aerobiosis enables S. cerevisiae to grow using fermentation products. This is not possible in

anaerobiosis due to the yeasts' inability to obtain energy from these products in the absence of oxygen. (iii) The rate of breakdown of sugar is similar in aerobiosis and in anaerobiosis; as fermentation is the prevalent reaction in both states, there is apparently no great difference in the ATP yield. In fact, the Pasteur effect in yeast occurs only under very special conditions such as when the cells are in the resting state or in sugar-limited-chemostat cultures. Surprisingly, it is generally believed that the 'Pasteur effect' is quantitatively important in *S. cerevisiae* and it has almost become a dogma that this phenomenon was first described by Pasteur in this microorganism.

XXI. Institute of Microbiology, Federal Institute of Technology, 8092 Zürich, Switzerland. Communicated by R. Hütter.

During this year two Ph.D. Theses have been finished in our yeast group. The summaries of these investigations follow below.

Kradolfer, P., Tryptophanstoffwechsel in *Saccharomyces cerevisiae*, Ph.D. Thesis ETH, Nr. 6769.

Saccharomyces cerevisiae degrades tryptophan very efficiently. The main degradation product is tryptophol. The initial step of the degradative pathway is catalyzed by two aromatic amino acid aminotransferases. The aromatic aminotransferase I is a constitutive enzyme. It catalyzes the aminotransfer from all aromatic amino acids to oxalacetate, 2-oxoglutarate, pyruvate and phenylpyruvate. The apparent K_m values are 6.10^{-3} M for tryptophan and 3.10^{-4} M for phenylalanine and tyrosine, respectively. The synthesis of the aromatic aminotransferase II has to be induced; all aromatic amino acids act as inducers which may serve as substrates as well. On the other hand, only pyruvate and phenylpyruvate can serve as aminoacceptors. The apparent K_m values are 4.10^{-4} M for tryptophan and 2.10^{-4} M for phenylalanine and tyrosine, respectively.

It could be shown that a loss of chorismate mutase activity causes a decrease of the feedback sensitivity of the anthranilate synthase. This decrease is most likely due to an increase of the chorismic acid pool. A strain lacking chorismate mutase activity, which is simultaneously constitutively derepressed and which possesses a feedback-resistant anthranilate synthase, accumulates anthranilic acid on all media tested. It is assumed that this accumulation reflects that the enzymes following anthranilate synthase work at their maximal level of 1.6 nmoles/min.mg protein. Most of the tryptophan synthesized is degraded immediately by the aromatic aminotransferases. A block of the aromatic aminotransferase II causes an increase of the tryptophan pool and of the tryptophan accumulation rate. The increase of the tryptophan pool leads to a tryptophan concentration high enough to allow the aromatic aminotransferase I to become active. Under optimal conditions, the tryptophan accumulation rate reached a level of 1.0 nmoles/min.mg protein.

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Stäheli, P., Biosynthese und Aminoacylierung von Transfer-RNA in *Saccharomyces cerevisiae*, Ph.D. Thesis ETH, Nr. 6843.

The role of transfer RNA (tRNA) in the regulatory system of Saccharomyces cerevisiae, called the general control of amino acid biosynthesis, was examined.

Growth inhibition, introduced by several amino acid limitations or by inhibition of the activity of tryptophanyl-tRNA synthetase, turned on the general control of amino acid biosynthesis. During limited amino acid growth conditions the cognate tRNAs were only poorly aminoacylated, thus indicating that decreased tRNA charging most likely was the primary signal of the general control of amino acid biosynthesis.

During tryptophan limited growth a new $tRNA^{Trp}$ isospecies accumulated that had an unmodified guanosine at position 17 of the primary sequence instead of the base 2'-O-methyl-guanosine. This incompletely modified $tRNA^{Trp}$ isospecies accumulated exclusively during growth conditions that prevented normal $tRNA^{Trp}$ charging. It was converted to the normal $tRNA^{Trp}$ species after supplementation of the growth medium with tryptophan. Most probably, the 2'-O-methylguanosine modification at position 17 could only be introduced after this molecule had been charged with tryptophan.

The incompletely modified $tRNA^{Trp}$ isospecies showed normal aminoacylation properties. Its charging level was decreased during tryptophan shortage to the same extent as that of the normal species. It is still unknown whether the incompletely modified $tRNA^{Trp}$ species plays a regulatory role in the general control of amino acid biosynthesis.

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Recently we published a paper on the "Biological role of the general control of amino acid biosynthesis" in Mol.Cell.Biol. 1, 584-593, 1981.

XXII. University of Göteborg, Department of Marine Microbiology, Carl Skottsbergs Gata 22, S-41319 Göteborg, Sweden. Communicated by Birgitta Norkrans.

Below follow abstracts of papers from our department, recently published, accepted or submitted for publication concerning halotolerance in yeast with Debaryomyces hansenii, isolated from marine environment, as the main test object; and metabolism of inorganic nitrogen in yeast with emphasis on hydroxylamine with Endomycopsis (= Saccharomycopsis) lipolytica as the main test object.

1. 1980 Adler, L. & Gustafsson, L.: Polyhydric alcohol production and intracellular amino acid pools in relation to halotolerance of the yeast Debaryomyces hansenii. - Arch. Microbiol., 124: 123-130.

   1981 Adler, L. & Liljenberg, C.: Sterol content, the fatty acid composition of phospholipids and permeability of labelled ethylene glycols in relation to salt tolerance of yeasts. Physiologia Plantarum. (In proof).

   1981 Adler, L., Falk, K.E., Norkrans, B. & Angström, J.: Glycerol and water in the salt tolerant yeast Debaryomyces hansenii as studied by nuclear magnetic resonance. FEMS Microbiol. Letters 11: 269-271.

1981 Lindman, B.: On the sugar uptake and halotolerance in the yeast Debaryomyces hansenii. Thesis, University of Göteborg ISBN 91-86022-06-7.

1981 Lindman, B.: D-glucosamine uptake by yeasts differing in halotolerance. FEMS Microbiol. Letters 10:379-382.

1981 Lindman, B.: Effect of NaCl on kinetics of D-glucosamine uptake in yeasts differing in halotolerance. Antonie van Leeuwenhoek 47:297-306.

1981 Lindman, B.: On the uptake of 2-deoxy-D-glucose and D-glucosamine by the halotolerant yeast Debaryomyces hansenii. Physiologia Plantarum (Accepted for publication).

1981 Lindman, B. & Norkrans, B.: Membranous interrelationships - an ultrastructural study of the halotolerant yeast Debaryomyces hansenii. Protoplasma (In proof).

1981 Lindman, B.: Influence of NaCl on the morphology and bud development of yeasts differing in halotolerance. (Submitted for publication).

The halotolerance of Debaryomyces hansenii has been explored previously in terms of its relation to high concentrations of NaCl as regards the respiration, the Na^+ and K^+ transport, alkaline phosphatase activity and energetic turnover and now as regards to intracellular polyols.

Two intracellular polyhydric alcohols, glycerol and arabinitol constitute the major osmotic components of this yeast during salt stress. There were no indications that amino acids played any role in osmoregulation. Findings in a study with HNMR technique applied on cells were interpreted to imply a significant restriction in motional freedom of the intracellular polyols due to interaction with macromolecular surfaces. Besides an osmoregulatory function the polyols might then serve as an agent for protecting the functional integrity of the biopolymers in the cell at low water activity.

To throw further light on the intrinsic halotolerance of D. hansenii studies were performed on (i) its sugar uptake both per se and in relation to NaCl, (ii) its ultrastructure and morphology.

The sugar uptake study was directed towards the glucose uptake by using the analogues glucosamine (slowly metabolisable) and 2-deoxy-glucose (non-metabolisable) applying radioassay technique. The comparison between D. hansenii and S. cerevisiae as regard the NaCl influence on the glucosamine accumulation unveiled a high capacity of the latter organism to transport sugar both in 0 M and 0.68 M NaCl as given by their kinetic parameters. However, the comparison between different salinities on an intra-species basis demonstrated an intrinsically halotolerant sugar transport in D. hansenii, since this yeast maintained its transport

capacity significantly better in increased salinity than did S. cerevisiae. Independent of the basis for forming the assessment, D. hansenii demonstrated also in 2.74 M NaCl an affinity for glucosamine maintained at a useful level.

The negative effect of anaerobiosis and metabolic inhibitors on the uptake of glucosamine and 2-deoxyglucose strongly suggests the operation of an active transport. Secondary active transport of sugar in form of a H^+ symport, often found in yeasts, however, could not be supported in D. hansenii; neither could the coupling of sugar uptake to the flux of Na^+ or K^+. Primarily a coupling to the antiport flux of anions therefore is suggested, although primary active transport must remain as an alternative.

The Woolf-Hofstee plots of glucosamine uptake in D. hansenii at different salinities may indicate that the negative effect of extreme salinity is exerted via the energy metabolism rather than directly to the transport apparatus per se.

The ultrastructure of D. hansenii, as revealed by transmission electron microscopy, displays an elaborate system of compartmentation structures. A variety of these structures, looking like and denoted as membranous protuberances, always originated from the plasma membrane. They could often be seen to extend into the vacuole. This frequent presence of osmiophilic structures suggests a significance in halotolerance, since they could provide a physiologically more optimized condition than would the cytoplasm. Their potentially high capacity of furnishing intracellular surfaces is likewise considered with respect to low water activity; enzymes attached to these surfaces may receive a better requisite for maintained activities.

Both the volume and shape were found to be influenced by increased substrate salinity. The observed volume reduction is suggested to be related to the concomitant decrease in specific growth rate, however, in a manner obscured by the presence of NaCl, since salt brings about more than pure starvation. The multifold effect of salt is emphasized in a tentative explanation inferring that e.g., the NaCl-inflicted metabolic inhibition is counteracted in a compensative enhancement of the size of the metabolic apparatus. This would involve an arrested cell volume decrease more pronounced in a more NaCl susceptible organism, such as S. cerevisiae. Bearing in mind the benefit of a larger cell surface to cell volume ratio, received automatically upon cell volume reduction, it is conspicuous that D. hansenii when adopting smaller cells grew simultaneously more elongated. The non-halotolerant S. cerevisiae rather grew more circular, thereby impairing its surface to volume ratio.

2. 1981 Norkrans, B & Tunblad-Johansson, I.: Changes in free amino acid content and activities of amination and transamination enzymes in yeasts grown on different inorganic nitrogen sources, including hydroxylamine. Antonie van Leeuwenhoek 47: 217-230.

1981 Tunblad-Johansson, I.: Metabolism of inorganic nitrogen in yeast with emphasis on hydroxylamine. - Thesis, University of Goteborg, ISBN 91-86022-05-9.

1981 Tunblad-Johansson, I. & Andersson, B.A.: Identification of 2-oximino acids in yeast by GC-MS. Acta Chem. Scand. (Accepted for publication).

The yeast, Endomycopsis lipolytica, has an exceptional ability to utilize hydroxylamine (HA) as the sole source of nitrogen. It was compared with the nitrate-utilizing yeast Cryptococcus albidus and with Saccharomyces cerevisiae, which requires completely reduced nitrogen for growth, with respect to intracellular metabolites and enzyme activities.

Gas chromatographic analysis of the content of Krebs cycle keto-acids in the three yeasts revealed a large α-keto-acid pool in E. lipolytica dominated by 2-oxoglutaric acid. The high level of 2-oxoglutaric acid was not only a response to hydroxylamine but a general characteristic for this yeast.

The free amino acids of the three yeasts were analysed as their n-propyl N-acetyl esters by gas chromatography. Special attention was paid to alanine, aspartic acid, and glutamic acid, the amino acids closely related to the Krebs cycle keto acids. The composition of the amino acid pool was similar for the three yeasts. Irrespective of nitrogen source glutamic acid was found as the principal amino acid rejecting the hypothesis that an obstructed amination could be responsible for the high level of 2-oxoglutaric acid in E. lipolytica. A positive correlation between the specific growth rate and the size of the amino acid pool was observed.

Assimilation of ammonia was mediated by NADP-linked glutamate dehydrogenase in all yeasts. Glutamine synthetase had an exclusively biosynthetic function since no activity of glutamate synthase was found in any yeast.

Formation of α-keto acid oximes may offer a way for microorganisms to utilize hydroxylamine. The oximes of pyruvic acid and 2-oxoglutaric acid were present in cells of HA-grown E. lipolytica and nitrate-grown Cr. albidus. No oxime was found in these yeasts grown on ammonia or in S. cerevisiae. These oximes could also be used as nitrogen sources for E. lipolytica.

The enrichment of ^{15}N in pyruvic acid oxime and free amino acids was determined by gas chromatography - mass spectrometry. The yeasts were suspended in media containing a ^{15}N-labelled nitrogen source. HA-suspended S. cerevisiae formed pyruvic acid oxime and a high ^{15}N-enrichment was observed but exhibited no incorporation of ^{15}N in amino acids. HA-suspended E. lipolytica displayed as high enrichment of ^{15}N in pyruvic acid oxime as S. cerevisiae but in this case ^{15}N was also incorporated in the amino acid pool of E. lipolytica.

XXIII. Instituto de Investigacion en Biologia Experimental, Facultad de Quimica, University of Guanajuato, Mexico. Communicated by J. Ruiz-Herrera.

The following is an abstract of a recent paper from our laboratory.

G. Larriba, M. Morales, and J. Ruiz-Herrera. 1981. Biosynthesis of β-Glucan Microfibrils by cell-free extracts from *Saccharomyces cerevisiae*. J. Gen. Microbiol. 124:375-383.

Glucan synthase activity in cell-free extracts of *Saccharomyces cerevisiae* was partially stabilized when cells were broken in the presence of sucrose. Under these conditions a significant amount of enzyme activity remained in the supernatant after high-speed centrifugation. When this supernatant fraction was incubated with UDPglucose, microfibrils were synthesized. Microfibrils were insoluble in water, ethanol and acid, and soluble in alkali. Under the electron microscope they appeared more or less uniform with an average length of about 0.5 μm. Alkali-insoluble residue appeared in the form of densely-packed longer microfibriles. After acidification of alkali-solubilized glucan, shorter microfibrils were reprecipitated. Microfibrils were digested by both endo- and exo-1,3-β-glucanase. In the latter case, glucose was the only product indicating that the microfibrils consist of 1,3-β-glucan with no detectable branches.

XXIV. Istituto di Microbiologia Agraria e Tecnica, University of Perugia, 06100 Perugia, Italy. Communicated by F. Federici.

The following papers have been submitted (or accepted) for publication:

Gabriella Monti and F. Federici. Extracellular Enzymatic Activities in the Yeast-Like Fungus *Aureobasidium pullulans*. Presented at the VIIIth Congress of European Mycologists, Bologna, Italy, 23-29 September 1981. An extended paper has been submitted to MYCOLOGIA.

Abstract

One-hundred and ninety-eight strains of the yeast-like microfungus *A. pullulans* were screened for their ability to release extracellular hydrolytic enzymes in the culture medium. All the isolates, or most of them, were found to be able to produce, though to varying extents, enzymes such as amylase, lipase (with different fats as substrates), pectinase (pectin-methylesterase and polygalacturonase), protease (as caseinolysis and gelatin liquefaction), nucleases (ribonuclease and deoxyribonuclease), urease and phosphatase. On the contrary, neither cellulase nor chitinase were produced. In some cases the activities, as detected in this preliminary screening, appeared to be interestingly high so the fungus can possibly be regarded as a promising source of extracellular lytic enzymes.

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G. Rosini, F. Federici and A. Martini. The Yeast Flora of Grape Berries During Ripening. Submitted for publication to MICROBIAL ECOLOGY

Abstract

The yeast flora associated with the surface of grapes during ripening was studied with regard to different sectors of the grapeskin and the position in the bunch by means of traditional as well as more vigorous pre-isolation and pre-counting treatments.

The yeast number per square cm of skin surface increases with ripening and is maximum in the area immediately surrounding the stem. The cluster sector closer to the peduncle seems to constitute a favourable substrate for yeasts, hosting a resident flora about ten and one hundred times higher than the central and lower parts of the bunch respectively. Kloeckera apiculata appears to be the normal resident species of grapes regardless of the sector or the ripening period and constitutes the fermenting flora of mature grapes. The ecological implications of the results of this survey are discussed.

* * *

F. Federici. A Note on the Milk-Clotting Ability in the Species of the Yeast Genera Cryptococcus and Rhodotorula. THE JOURNAL OF APPLIED BACTERIOLOGY (in press).

Summary

One-hundred and forty-five yeast strains, 84 belonging to the genus Cryptococcus and 61 to the genus Rhodotorula, were screened for their milk-clotting ability. All the Rhodotorula and most of the Cryptococcus species showed a significant milk coagulating capacity. The results of the present study could possibly be of some taxonomic value and, furthermore, certain isolates seem interesting as producers of extracellular protease.

XXV. Institut für Mikrobiologie Und Weinforschung, Johannes Gutenberg-Universität, Ernst-Ludwig-Strasse 10, 6500 Mainz, West Germany. Communicated by F. Radler.

Below follow the summaries of two recent papers from our Institute.

Luitgard Reinhard und Ferdinand Radler. Die Wirkung von Sorbinsäure auf Saccharomyces cerevisiae. I. Beeinflussung des Wachstums sowie der aeroben und anaeroben Glucoseverwertung. Zeitschrift für Lebensmittel-Untersuchung und-Forschung 172:278-283 (1981).

Summary

Sorbic acid inhibits the growth of yeasts and leads to the death of the cells. The degree of effect of sorbic acid depends on its concentration, the culture condition, and the number of yeast cells present. Growth and metabolism of yeast are influenced to varying degrees. Low concentrations of sorbic acid (50-100 mg/l) cause a significant inhibition of the growth and multiplication of yeast cells, without showing an influence on fermentation. Yeast cells that have been exposed for a short time to concentrations of 500 to 1,500 mg/l sorbic acid are killed and therefore unable to form colonies on sorbic acid free growth media. The fermentation and respiration of resting yeast cells is much less influenced by sorbic acid than cell growth. It was demonstrated that the total amounts of CO_2 and ethanol produced by yeast cells were not affected by concentrations of up to 500 mg/l sorbic acid at pH 4.0. The formation of CO_2 from glucose by cell free extracts of Saccharomyces cerevisiae was not inhibited by 3,000 mg/l sorbic acid. Similarly the

enzymes aldolase, enolase and pyruvate decarboxylase were not inhibited by high concentrations of sorbic acid. Therefore it is assumed that sorbic acid acts on the yeast cell mainly by influencing its cell membrane and its permeability.

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Luitgard Reinhard und Ferdinand Radler. Die Wirkung von Sorbinsäure auf Saccharomyces cerevisiae. II. Adsorption, Desorption und Abbau von Sorbinsäure. Zeitschrift fur Lebensmittel-Untersuchung und-Forschung 172:382-388 (1981).

Summary

Cells of Saccharomyces cerevisiae adsorb sorbic acid within a brief period. The amount of acid adsorbed depends on the yeast cell mass and sorbic acid concentration. As much more sorbic acid is adsorbed at pH 3 than at pH 7 it may be assumed that it is the undissociated molecules that are adsorbed by the yeast cells. However, no accumulation of sorbic acid is observed within the yeast cells, for the acid is quantitatively present in the medium after an incubation of several days, if no degradation has occurred. The concentration of sorbic acid is greatly increased within the space surrounding the cells by adsorption; this is most likely of importance for the inhibition of cell growth. A small part of the sorbic acid is decomposed by the yeast cells, independent of the pH of the medium. The mode of degradation is unknown, but this part of sorbic acid is converted into a form that cannot be analyzed by UV-absorption. Yeast cells growing in the presence of sorbic acid (100 mg/l) tend to produce pseudomycelium. It is theorized that the disturbance of cell growth may be caused by a reaction of sorbic acid with thiol groups of the surface of the yeast cell.

XXVI. Alko, Box 350; SF-00101 Helsinki 10, Finland. Communicated by Heikki Suomalainen.

Below follow abstracts of work published since June 1981.

Heikki Suomalainen. Yeast Esterases and Aroma Esters in Alcoholic Beverages. Journal of the Institute of Brewing 87: 296-300 (1981).

It has been shown that the proportion of fatty acid ethyl esters retained by the yeast cell increases with increasing acyl chain length as the ester becomes more lipid soluble. The distribution of esters depends on the yeast strain and on the fermentation temperature; larger amounts of esters were found to transfer from the cells into the medium at higher temperature. It was shown that esterase activity is located both inside and outside the yeast cell plasma membrane. Intact yeast was capable of hydrolysing the ethyl esters of caproic, caprylic and capric acid. Acetate esters were hydrolysed only very slowly or not at all.

The hydrolytic activity of baker's yeast was studied with ethyl caprylate as substrate. The hydrolysis was very fast at the beginning. The equilibrium attained depended not only on the concentration of ester and alcohol but also on the pH, a higher amount of ester remaining in

solutions of lower pH. It was also shown that the esterases possess appreciable ester synthesizing ability and an equilibrium was attained by incubating yeast with caprylic acid and ethanol.

The experiments described show that the ester level in an alcoholic beverage, such as beer, is not dependent solely on the ester concentration formed during fermentation: in the presence of yeast the level can be shifted in either direction by changing temperature, pH or alcohol concentration - or the amount and type of yeast.

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I. Molnar, E. Oura and H. Suomalainen. Changes in the Activities of Certain Enzymes of Champagne Yeast during Storage of Sparkling Wine. Acta Alimentaria Academiae Scientiarum Hungaricae 9: 313-324 (1980).

Active and NaF-inhibited Champagne Hautvilliers champagne yeast were added to tank-fermented sparkling wine and the activities of the following yeast enzymes were serially measured during storage at different temperatures: alcohol dehydrogenase, aldolase, esterase, glucose-6-phosphate dehydrogenase, glycerolaldehyde-3-phosphate dehydrogenase, glutamate-oxalacetate transaminase, hexokinase, β-fructosidase, malate dehydrogenase and protease. On the basis of the changes in activity, the enzymes could be divided into three groups. Alcohol dehydrogenase, aldolase and glutamate-oxalacetate transaminase lost their activities most rapidly; the activities of glycerolaldehyde-3-phosphate dehydrogenase, hexokinase and malate dehydrogenase were still considerable after a storage of 50-90 days, depending on the temperature; the β-fructosidase activity showed hardly any change, and the proteolytic activity increased, during storage. The length of the period in which, following the second fermentation, enzyme activity is still considerable is estimated between 30 and 60 days.

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I. Molnar, E. Oura and H. Suomalainen. Determination of the Autolysis of Champagne Yeast by Using ^{14}C-labelled Yeast. Acta Alimentaria Academiae Scientiarum Hungaricae 9: 305-312 (1980).

The degree of autolysis of ^{14}C-labelled Champagne Hautvillers yeast was followed up at different temperatures of storage. A linear relationship was found between the length of the storage and the degree of autolysis. The rate of autolysis increased with raising the temperature of storage. Raising of the temperature by 10°C was followed by a 6-7% increase in the rate of autolysis. Shaking up the yeast sediment at 20-day intervals raised the rate of autolysis by 1.5-4.2%.

* * *

Sampsa Haarasilta. On the Regulation of Anaplerotic and Gluconeogenic Enzymes in Baker's Yeast (Saccharomyces cerevisiae). Thesis, Helsinki University, 1981, 72 pp.

The investigation deals with the activity and regulation of anaplerotic and gluconeogenic enzymes in yeast cells. The anaplerotic

enzymes have a central importance for the production of cell mass and metabolites from non-sugar nutrients such as ethanol.

The amounts of anaplerotic and gluconeogenic enzymes were found to be much dependent on the composition and aeration of the growth solution. Important factors were e.g., its main nutritive source and the biotin content was well as the amount of available oxygen. Tracer studies with radioactive carbon dioxide, however, indicated that the amount of enzymes is not the only factor that regulates the anaplerotic and gluconeogenic reaction paths during growth.

When the enzyme determination procedures were developed, a very sensitive and specific method for determining biotin (H vitamin) was also developed by which picogram amounts of biotin can be detected.

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The following publication has appeared since the last communication. The abstract of report has been given in Yeast Newsletter 27(1978): 2, 88-89.

E. Oura. Estimation of the flux of the substrate at different steps of metabolism during microbial growth. In: "Continuous Cultivation of Microorganisms", Proc. Symp., 7th, Prague 1978, ed. by B. Sikyta, Z. Fencl and V. Polacek, Praha 1980, pp. 91-100.

XXVII. Ministère de L'Agriculture, Ecole Nationale Supérieure des Industries Agricoles et Alimentaires Centre de Douai, 13, Rue de l'Université, 59509, Douai, France. Communicated by M. Bouix.

Below follow titles and abstracts of recent works from this research station.

M. Bouix and J.Y Leveau. Detection of Wild Yeasts in Brewing Yeasts by Double Fluorescence. MBAA Technical Quarterly, 17:163-166 (1980).

Abstract

For the detection of wild yeasts in brewing yeasts, a double fluorescence technique was applied. We studied the possibility of preparing a specific anti yeast culture serum. Since *Saccharomyces carlsbergensis* cells are rendered specifically fluorescent by fluorescein isothiocyanate, we stained yeast contaminants with another fluorochrome: Evans blue. The *S. carlsbergensis* brewing yeast cells visualized with this technique had fluorescent green cell walls and reddish brown cytoplasm, whereas contaminant yeast cells were a clear red. We tested this technique on 40 yeast strains of different genera and species. On these 40 yeast strains tested, we were thus able to distinguish 37 from the brewing yeast. These results show that this double fluorescence technique may be applied to rapid detection of brewery contaminants, but that it is not 100 percent reliable because of the serological variations within the same species.

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M. Bouix, J.Y Leveau and C. Cuinier. Determination de L'Origine des Levures de Vinification par Une Méthode de Differenciation Fine des Souches. Connaissance de la Vigne et du Vin, 15:41-51 (1981).

Summary

The authors compare cultures of Saccharomyces cerevisiae by electrophoresis of their endocellular parts. The cultures were isolated during winemaking, on the products involved from the vine to the wine, and at different points of contamination. This technique allows to finely differentiate yeast cultures of the same species.

It appears that several clones of S. cerevisiae are present from harvest until pressing, as much on the products as on the harvesters hands and harvesting materials, while during fermentation, only one clone was isolated. The presence of this clone was found from the first stages of vinification, but has not been isolated in the winery.

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M. Bouix, J.Y. Leveau and C. Cuinier. Reconnaissance des Souches de Levures de Même Espèce. Colloque Soc. Fr. Microbiol., Reims, 1981, pp. 235-249.

XXVIII. Agricultural Microbiology Section, Dairy and Food Science Department, University of Saskatchewan, Saskatoon, Canada S7N 0W0. Communicated by W.M. Ingledew.

The following papers have been published:

W.M. Ingledew, J.D. Burton, D.W. Hysert, G. Van Gheluwe. Membrane Filtration: Survival of Brewing Microbes on the Membrane During Storage at Reduced Humidities. J. Amer. Soc. Brew. Chem. 38(4):125, 1980.

Abstract

The membrane filtration technique as applied in multiplant brewery quality control has been unsatisfactory because of dehydration and subsequent death of entrapped microbes during shipment (storage) of membranes. Gram-negative bacteria and lager yeast were shown to be very susceptible to such storage over five days at humidities ranging from 0 to 98%. Gram-positive brewing bacteria, however, were much more resistant over the whole range of humidity. A number of compounds were used as protective agents in an attempt to prevent dehydration and subsequent death. One of them, 4% reconstituted skim milk powder, was extremely effective in reduction of death and is now recommended as a protective washing solution for microbes collected on membranes. Use of this protective agent allows membranes to be shipped by mail in sterile Whirl pak bags under a variety of conditions with no appreciable microbial die-off.

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W.M. Ingledew, J.D. Burton.
Samplers for Dip Testing or Swab Testing in Breweries. J. Amer. Soc. Brew. Chem. 39(1):39, 1981.

Abstract:

Experiments with Millipore Yeast and Mold and Total Count TM samplers have demonstrated that recovery of typical brewery bacteria and yeasts from suspensions is poorer when samplers are used than when traditional membrane filtration is used. Based on these data, such samplers, at least in their present mode, cannot be recommended for use in the brewery.

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W.M. Ingledew, G.A. Jones
The Fate of Live Brewers' Yeast Slurry in Bovine Rumen Fluid. J. Inst. Brewing 87: , 1981 (in press).

Abstract

Live brewers' yeast slurry was incubated under carbon dioxide at 27°C and 39°C in 0.1% peptone solution and in bovine rumen fluid which had been clarified by removal of the population of bacteria and protozoa normally present. Numbers of viable yeast in both media remained constant for 12 h at 27°C; at 39°C loss in viability was 81% in peptone and 94% in rumen fluid during the same period. When glucose was added to clarified or unclarified rumen fluid containing yeast slurry and incubated for 6 h at 39°C ethanol was produced. Ethanol production was prevented if the slurry was treated with heat or chemical preservatives before addition to the rumen fluid. Unclarified rumen fluid from a steer fed a brome-alfalfa hay-grain ration contained 10^2-10^3 yeasts and moulds per ml. The results suggested that the feeding of live brewer's yeast slurry to ruminants could result in ethanol toxicity if fermentable carbohydrate were also present, though many of the yeast cells would succumb to heat inactivation at normal rumen temperatures. This risk could be eliminated by prior treatment of the slurry with heat or chemical preservatives.

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E.W.T. Tsang, W.M. Ingledew
Studies on the Heat Resistance of Wild Yeasts and Bacteria in Beer. J. Amer. Soc. Brew. Chem. 39(4): , (in press).

Abstract

Detailed kinetic studies on wild yeast and heat-resistant brewing bacteria showed that the Z value used in current lethal-rate calculations in brewing is not suitable. An alternative method, based on the D values of the most heat-resistant brewing microbe, is proposed. A complete reinvestigation of pasteurization in brewing is suggested.

XXIX. Research Institute for Viticulture and Enology, Matuskova 25, 886 15 Bratislava, Czechosolvakia. Communicated by E. Minarik

The following is the summary of a paper accepted for publication in Kvasny prumysl (Fermentation Industry) Prague in 1982:

E. Minarik: Possibilities of influencing grape must fermentation by the activator of Botrytis cinerea (in Slovak).

The influence of the activator originating from the hyphal fungus Botrytis cinerea Pers. on the fermentation rate and reproduction ability of Saccharomyces oviformis and on the composition of the fermented substrate has been examined under laboratory conditions. Already by minimal doses of the dried mycelial preparation of B. cinerea a remarkable acceleration of the fermentation rate, a more effective sugar utilization, higher alcohol content and a more favourable composition of the grape wine could be achieved. Possible technological conclusions in the production of natural sweet wines are discussed.

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In Paris the 16th Annual Session of the Subcommission "Microbiology of Wine" of the Office International de la Vigne et du Vin was held on May 20, 1981. The following main papers were presented:

S. Lafon-Lafourcade (University of Bordeaux II): Vinegar bacteria of Wine.

F. Vezinhet (I.N.R.A., Montpellier): Application of Genetics in Winemaking. Methodology and Objectives.

Ch. Divies (University of Dijon): Possibilities of fixed enzymes and microorganisms: application in winemaking.

E. Minarik (CSSR): Practical methods of examination of enological properties of yeasts - remarks and discussion to 9 proposed methods.

* * *

E. Minarik: Ecology of yeasts and yeast-like microorganisms of barrel and bottled wines. Trilateral Conference on Yeasts. Horticultural University, Budapest, July 27-31, 1981.

Problems connected with the occurrence of the osmotolerant, chemoresistant and fructophilic yeast Saccharomyces bailii var. bailii were discussed and measures to prevent wine spoilage and haze and secondary fermentation of sweet wines, respectively, presented.

XXX. Meetings

1. International Commission for Yeasts. September 24, 1981. Held at the University of Valencia, Valencia, Spain.

Members present at the September 24, 1981 meeting of the ICY in Valencia, Spain.

(Belgium) H. Verachtert; (Canada) B. Johnson, G. Stewart; (Czechoslovakia) P. Biely for A. Kockova-Kratochvilova, K. Sigler for A. Kotyk; (Finland) M. Korhola for H. Suomalainen; (France) J. Bastide; (Great Britain) A.H. Rose; (Japan) T. Hirano for S. Nagai; (Portugal) N. van Uden; (Scotland) L. Douglas for I. Dawes; (Spain) J. Gancedo, R. Sentandreu; (Switzerland) O. Kappeli; (U.S.S.R.) N. Elinov.

The following members sent their apologies for absence.

(Austria) H. Klaushofer, (Brazil) A. Panek, (Canada) I. Russell, (Czechoslovakia) A. Kockova-Dratochvilova, (Czechoslovakia) A. Kotyk, (Czechoslovakia) E. Minarik, (Denmark) A. Stenderup, (England) J. Spencer, (Finland) H. Suomalainen, (France) H. Heslot, (India) T. Subbaiah, (Ireland) A. Forage, (Japan) S. Nagai, (Netherlands) J. Hoogerheide, (Netherlands) L. Rodrigues de Miranda, (Poland) J. Jakubowska, (U.S.A.) H.J. Phaff and F. Sherman, (East Germany) H. Koch, (West Germany) S. Windisch, (Yugoslavia) V. Johanides.

Membership of ICY

Dr. S. Windisch sent a letter of resignation as West Germany's representative on the ICY and Professor C. Emeis was elected in his place. The Commission thanked Professor Windisch for his years of service and sent a letter to him with those sentiments.

Dr. J. Hoogerheide also sent a letter of resignation and the Commission sent a letter thanking him for his years of service.

The Following New Members Were Elected

A. Forage (Ireland)
N. van Uden (Portugal)
O. Kappeli (Switzerland)
C. Emeis (West Germany)
T. Lachowicz (Poland)

IUMS

The Chairman reported that the International Union of Microbiological Societies (IUMS) has now been formed out of the old International Association of Microbiology Societies (IAMS).

A letter was sent to Dr. Kazuo Iwata, on behalf of the ICY, stating that this Commission wishes to be affiliated with the Mycology Division of the International Union of Microbiological Societies (IUMS).

VIth ISY

Professor A.H. Rose announced that the VIth ISY will be held in Bath, England commencing on September 19, 1984 and would last for about 3-1/2 days. Professor Rose stated that a British

organizing committee had been established and that further details would be available at the next commission meeting.

The first circular for the VIth ISY would be mailed in late 1982.

The present thinking was that certain areas (for example, Pathogenic Yeasts) would attract special attention and between 350 to 400 people will probably be attending.

Future ISSY Meetings

VIIIth Specialized Symposium on Yeast (ISSY)
January 17th, 1983 - Bombay, India
"Yeast Technology" (Dr. T.V. Subbaiah)

IXth Specialized Symposium on Yeast (ISSY)
Late Spring or early Summer 1983 - Smolence Castle, Czechoslovakia
"Yeast in the Human Environment" (Dr. A. Kockova-Kratochvilova)

These was some discussion regarding the availability of space in Smolence Castle and the Czechoslovak delegates undertook to investigate this matter and report to the Chairman.

Xth Specialized Symposium on Yeast (ISSY)
Mid 1985 - Sofia, Bulgaria
"Molecular and Genetic Aspects of Yeast" (Dr. P.V. Venkov)

XIth Specialized Symposium on Yeast (ISSY)
1986 - Portugal
"Basic Research on Applied Aspects of Yeast" (for example, drug action, influence of ethanol and other primary metabolites of yeast, the genetics of yeast with respect to ethanol and other drugs, etc).

Yeast Newsletter

In the absence of Professor H.J. Phaff, the Chairman reported that Professor Phaff will continue acting as editor of the Yeast Newsletter for at least the next two to three years.

Date/Place of Next Meeting

This was left at the call of the chair but it was agreed that if 10 or more Commission members attended the XIII International Congress of Microbiology (ICM) in Boston (August 1982) an ICY meeting could be held at that time.

In closing, the Chairman expressed, on behalf of all commission members, their gratitude to Professor Sentandreu and the organizing committee for the successful symposium held in Valencia, Spain.

There being no other business, the meeting was adjourned.

G.G. Stewart
Chairman - ICY

2. The Fourteenth Annual Meeting of the Yeast Genetics Conference - Japan was held from August 24 to 26, 1981, at Tanaka Memorial Hall of Osaka City University, Osaka. The following topics were presented and discussed.

Session 1: Structure and Function of Cell Organelles (Chairpersons, H. Mori, K. Tanaka, H. Tamaki and T. Hirano).

Yoshio Tani, Yukiko Yamada and Teijiro Kamihara* (Seibo Women's Jr. Col., *Dept. Ind. chem., Kyoto Univ.). Effects of polyamines on cell morphology of yeasts.

Teijiro Kamihara, Ichiro Nakamura, Norihisa Nakamura, Michio Nakai, Saburo Fukui, Masako Osumi* and Eiichi Nakai** (Dept. Ind. chem., Kyoto Univ., *Dept. Biol., Japan Women's Univ., **Fac. Med., Kyoto Univ.). Action of vitamin B_1 on yeasts: Morphology and functions of mitochondria.

Takushi Hatano, Eiko Tsuchiya, Tokichi Miyakawa and Sakuzo Fukui (Dept. Ferment. Technol., Hiroshima Univ.). Histochemical study on morphogenesis of heterobasidiomycetous yeasts.

Kanji Takeo and Ei-ichi Nakai* (Res. Inst. Chemobiodynamics, Chiba Univ., *Fac. Med., Kyoto Univ.). The plasma membrane of Pityrosporum has natural markers to show how the cell grows II.

Misuzu Nagano and Masako Osumi (Dept. Biol., Japan Women's Univ.). Ultrastructure of the yeast cell by rapid freezing.

Masako Osumi and Naoko Yamada* (Dept. Biol. and *Lab. Electron Microscopy, Japan Women's Univ.). The three-dimensional structure of the crystalloid yeast microbody.

Junji Watari, Takashi Sasaki and Norio Nishikawa (Res. Labs., Sapporo Breweries Ltd.). Preservation of yeast spheroplasts.

Tadashi Hirano, Akira Tanaka* and Yoshio Sekiguchi* (Jikei Univ. Sch. Med., ctr. Res. Lab., *Tokyo Metro. Inst. Med. Sci.). Visualization of fusion and regeneration of yeast protoplasts.

Kenji Doi (Inst. Sci. Ind. Res., Osaka Univ.). Mode of progress of mitotic cycle of yeast spheroplasts in liquid culture.

Yasuo Nakatomi (Oriental Yeast Co. Ltd.). Protoplast fusion between Saccharomyces rosei and Saccharomyces cerevisiae.

Hideo Tamaki (Doshisha Women's College). Genetic character of abortive cells on protoplast fusion in yeasts.

Session 2: Radiation and Mutation (Chairpersons, K. Hieda and M. Yanagida).

Katsumi Kobayashi (Inst. Biol. Sci., Univ. Tsukuba). Biphasic induction of gene conversion in proton-bombarded yeast, *Saccharomyces cerevisiae*.

Kazuo Shindo and Jumpei Amagasa (Dept. Phys., Rikkyo Univ.). Binding of acridine orange to yeast ribosomes.

Yoshihiko Hayakawa and Kotaro Hieda (Dept. Phys., Rikkyo Univ.). Effects of UV dose fractionation on survival and gene conversion in *Saccharomyces cerevisiae*.

Yumiko Ishino and Bun-ichiro Ono (Dept. Pharm. Technol., Okayama Univ.). Arginine uptake of a suppressed canavanine-resistant mutant.

Bun-ichiro Ono, Manli Weng* and Kezhong Tong* (Dept. Pharm. Technol., Okayama Univ., * Inst. Genetics, Acad. Sinica). Genetic analysis of chromium-resistant and sensitive mutants in *Saccaromyces cerevisiae*.

Session 3: Cytoplasmic Inheritance (Chairperson, K. Suda).

Fumiko Miyamoto (Dept. biol., Wakayama Univ.). Effect of mono- and di- nitrophenols on growth and respiratory deficient mutation in yeast.

Yoshihisa Iwamoto and K. Lemone Yielding* (Dept. Microbiol., Shizuoka Col. Pharm., * Dept. Anat., Univ. South Alabama). Induction of sector mutants of *Saccharomyces cerevisiae* DP1 1B/517 by propidium.

Susumu Nagai, Sayoko Ochi and Kimiomi Nishimura (Dept. Biol., Nara Women's Univ.). Lactose fermentation, resistance to cycloheximide, and respiration deficiency in *Kluyveromyces marxianus* (A preliminary report).

Masaaki Kitano and Tetsuo Murayama (Biol. Inst., Ehime Univ.). Genetic analysis of the cadmium-resistant yeast II.

Session 4: Gene Regulation, Recombination and Mapping (Chairpersons, I. Takano, K. Matsumoto, B. Ono and S. Okamoto).

Tadanori Yoshimatsu and Kunihiro Matsumoto (Dept. Ind. Chem., Tottori Univ.). Isolation and characterization of mutations resistant to catabolite repression in *Saccharomyces cerevisiae*.

Kunihiro Matsumoto and Isao Uno* (Dept. Ind. Chem., Tottori Univ., * Inst. Appl. Microbiol., Univ. Tokyo). The role of cyclic AMP and cyclic AMP-dependent protein kinase in *Saccharomyces cerevisiae*.

Isao Uno and Kunihiro Matsumoto* (Inst. Appl. Microbiol., Univ. Tokyo, * Dept. Ind. Chem., Tottori Univ.). The role of cyclic AMP and cAMP-dependent protein kinase in Saccharomyces cerevisiae.

Junpei Ishiguro and Yutaka Arakatsu (Dept. Biol., Konan Univ.). Genetic and biochemical characterization of yeast antisuppressor mutants.

Jiro Akimaru, Yoshinobu Kaneko, Akio Toh-E* and Yasuji Oshima (Dept. Ferment. Technol., Osaka Univ., * Dept. Ferment. Technol., Hiroshima Univ.). Was PHO83 mutation caused by insertion of Tyl?

Kazuma Tanaka, Satoshi Harashima and Yasuji Oshima (Dept. Ferment. Technol., Osaka Univ.). Mating-type dependent homozygosity of sir3 mutation in yeast.

Takashi Toda, Kazuhiko Umesono, Masayuki Yamamoto* and Mitsuhiro Yanagida (Dept. Biophys., Kyoto Univ., * Inst. Med. Sci., Univ. Tokyo). Genetic analysis and mapping of cold-sensitive cell division cycle (cs cdc) mutations in Schizosaccharomyces pombe.

Kazuhiko Umesono, Takashi Toda and Mitsuhiro Yanagida (Dept. Biophys., Kyoto Univ.). Genetic analysis of a cs cdc mutant supersensitive to antimitotic benzimidazole compounds in Schizosaccharomyces pombe.

Kazuhisa Sakai and Toshiaki Takahashi (Dept. Microbiol. Centr. Res. Lab., Asahi Breweries Ltd.). Genetic analysis of genes controlling H_2S production in Saccharomyces cerevisiae.

Michio Tsuboi (Dept. Biol., Osaka City Univ.). Genetic analysis of sporulation-deficient mutants in Saccharomyces cerevisiae.

Toyohiko Yamazaki (Dept. Ferment. Technol., Yamanashi Univ.). Sequence of the genes on chromosome III in Saccharomycodes ludwigii.

Session 5: Plasmid and Cloning (Chairpersons, M. Takagi, T. Gunge and M. Yamamoto).

Norio Gunge (Mitsubishi-Kasei Inst. Life Sci.). Intergeneric transfer of pGKl killer plasmids from Kluyveromyces lactis into Saccharomyces cerevisiae.

Atsuko Takagi, Satoshi Harashima and Yasuji Oshima (Dept. Ferment. Technol., Osaka Univ.). Yeast transformation is highly associated with cell fusion.

Yoshinobu Kaneko, Akio Toh-E* and Yasuji Oshima (dept. Ferment. Technol., Osaka Univ., * Dept. Ferment. Technol., Hiroshima Univ.). Cloning of two DNA sequences which complement repressible alkaline phosphatase negative (pho9) mutation.

Masahide Kawamura, Masamichi Takagi and Keiji Yano (Dept. Agric. Chem., Univ. Tokyo). Cloning of a *Candida* gene in *Escherichia coli*.

Masayuki Yamamoto and Junko Sakaguchi (Inst. Med. Sci., Univ. Tokyo). Cloning and structure analysis of a fission yeast *ura* gene.

Toshimichi Ikemura (Dept. Biophys., Kyoto Univ.). Choice of codon in yeast genes is largely constrained by the availability of transfer RNAs.

Session 6: Biochemistry (Chairpersons, T. Mizunaga and N. Sando).

Etsuko Uchida, Yoshinori Ohsumi* and Yasuhiro Anraku* (Dept. Biol., Japan Women's Univ., * Dept. Biol., Univ. Tokyo). Plasma membrane Mg^{2+}-ATPase of *Saccharomyces cerevisiae*.

Yoshinori Ohsumi and Yasuhiro Anraku (Dept. Biol., Univ. Tokyo). Ca^{2+} transport system of vacuoles from *Saccharomyces cerevisiae*.

Masazumi Miyazaki (Inst. Mol. Biol., Nagoya Univ.). Studies on a peptide chain elongation factor, EF-3, characteristic for yeast.

Shun Okada and Mitsuhiro Yanagida (Dept. Biophys., Kyoto Univ.). DNA topoisomerase activities in isolated nuclei of *S. pombe*.

Toshihiro Noguchi, Takemitsu Mizunaga, Yoshiharu Maruyama (Dept. Agr. Chem., Univ. Tokyo). Immunological evidence for the accumulation of non-glycosylated acid phosphatase peptides in the membrane fraction of yeast protoplasts treated with tunicamycin.

Tetsuyoshi Suizu, Eiko Tsuchiya, Tokichi Miyakawa and Sakuzo Fukui (Dept. Ferment. Technol., Hiroshima Univ.). Alteration in molecular pattern of nuclear proteins causing by progression of cell division cycle in *Saccharomyces cerevisiae*.

Kazuo Kamimura, Eiko Tsuchiya, Tokichi Miyakawa and Sakuzo Fukui (Dept. Ferment. Technol., Hiroshima Univ.). Cytoplasmic factor controlling DNA synthesis in isolated nuclei of *Saccharomyces cerevisiae*.

Session 7: Sporulation and Life Cycle (Chairpersons, S. Harashima, M. Osumi, T. Simoda and M. Tsuboi).

Katsuji Ueki and Nobundo Sando* (Fac. Agric., Yamagata Univ., * Fac. Sci., Yamaguchi Univ.). Physiological changes during sporulation in *Saccharomyces cerevisiae*.

Michio Tsuboi and Masako Ohi (Dept. Biol., Osaka City Univ.). Analysis of sporulation by protoplast-fusion technique.

Chikashi Shimoda and Masaaki Tanaka (Dept. Biol., Osaka City Univ.). Control of trehalase activity during sporulation in the yeast Schizosaccharomyces pombe.

Aiko Hirata and Kenji Tanaka* (Inst. Appl. Microbiol., Univ. Tokyo, * Nagoya Univ. Med.). Fine structure of ascospore formation in the fission yeast, Schizosaccharomyces.

Nobundo Sando, Isamu Miyakawa, Tohru Eguchi, Sohryu Nishibayashi* and Tsuneyoshi Kuroiwa* (Fac. Sci., Yamaguchi Univ., * Dept. Cell Biol., Natl. Inst. Basic Biol.). Arrangement of mitochondrial nucleoids during meiosis and sporulation in Saccharomyces cerevisiae.

Susumu Okamoto (Dept. Biol., Univ. Tokyo). A positive regulator for spindle pole body morphogenesis at the second meiotic division in Saccharomyces cerevisiae.

Naohiko Yanagishima, Hiroaki Fujimura and Isao Banno* (Biol. Inst., Nagoya Univ., * Inst. Ferment., Osaka). Sex pheromones in ascosporogenous yeasts.

Hiroaki Fujimura, Kazuo Yoshida and Naohiko Yanagishima (Biol. Inst., Nagoya Univ.). Purification and characterization of binding substance (BS) to α pheromone in Saccharomyces cerevisiae.

Hiroshi Tohoyama and Naohiko Yanagishima* (Biol. Inst., Ehime Univ., * Biol. Inst., Nagoya Univ.). Control of the sexual agglutinability by the mating type locus in Saccharomyces cerevisiae.

Yoshiyuki Nakagawa and Naohiko Yanagishima (Biol. Inst., Nagoya Univ.). Sexual agglutinability and cell membrane.

Satoshi Hasegawa and Naohiko Yanagishima (Biol. Inst. Nagoya Univ.). Differential inhibition of induction of sexual agglutinability and zygote formation.

Makoto Yamaguchi, Kazuo Yoshida and Naohiko Yanagishima (Biol. Inst., Nagoya Univ.). Purification and partial characterization of agglutination substance from the cytoplasm in the yeast Saccharomyces cerevisiae.

Session 8: Reports (Chairperson, S. Nagai).

Masayuki Yamamoto (Inst. Med. Sci., Univ. Tokyo). Reports of the 1981 Cold Spring Harbor Molecular Biology Yeast Meeting.

Communicated by Kotaro Hieda
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Biophysics Laboratory
Nishi-Ikebukuro 3, Toshima,
Tokyo 171, Japan

3. The Gulbenkian Institute of Science, Oeiras, Portugal will offer in 1982 the following two international courses on yeasts:

30 August - 10 September. Effects of Alcohols and Other Membrane-Active Drugs on Yeast and Other Microorganisms. Anthony H. Rose (University of Bath, U.K.), L.O. Ingram (University of Florida, Gainesville), Vitor M.C. Madeira (University of Coimbra), N. van Uden (Gulbenkian Institute of Science, Portugal).

20 September - 15 October. Taxonomy and Identification of Yeasts. N.J.W. Kreger-van Rij (University of Groningen, Netherlands), C.P. Kurtzman (NRRC, Peoria, Illinois), J. Fell (University of Miami, Florida), S.A. Meyer (Georgia State University, Atlanta).

Requests for information and application for admission forms should be directed to:

Prof. N. van Uden
Laboratory of Microbiology
Gulbenkian Institute of Science
2781 Oeiras Codex
Portugal

4. International Course on Biochemistry and genetics of Yeasts, July 5-24, 1982 Madrid, Spain (sponsored by FEBS and ICRO). The purpose of the course is to acquaint the participants with the potentialities of yeast as a model eukaryotic system to study enzyme regulation and gene expression and to introduce them to the basic research methodologies in the biochemistry and genetics of yeast.

Information: Dr. C. Gancedo
Instituto de Enzimologia del C.S.I.C.
Facultad de Medicina, Universidad Autonoma
Arzobispo Morcillo, 6, Madrid-34, Spain

Deadline: 31 March 1981

5. The XIth International Conference on Yeast Genetics and Molecular Biology will be held in Montpellier from the 13th to the 17th of September 1982.

The organizers are P. Galzy (Montpellier) and P. Pajot (Gif-sur-Yvette). P.P. Slonimski is in charge of the Scientific Program.

This first announcement is sent to the participants to the Tenth International conference in Louvain. Please forward the information to those interested, or let the organizers know their names and addresses.

Further information will be forwarded in a few months to those who indicated their interest by filling out and sending the following form to:

XIth International Yeast Conference
Chaire de Genetique et Microbiologie
ENSAM Place Viala
34060 Montpellier Cedex, France

I am interested in further information concerning the XIth International Conference on Yeast Genetics and Molecular Biology

NAME (Capitals):

ADDRESS (Capitals):

6. Third International Mycological Congress (IMC), Tokyo, Japan, 28 August - 3 September 1983. The first circular may be obtained from Professor K. Tubaki at the address below:

Prof. K. Tubaki
Secretary General
Third International Mycological Congress
c/o International Congress Service, Inc.
Chikusen Bldg. 5F
2-7-4, Nihombashi
Chuo-ku
Tokyo
Japan 103

XXXI. Brief News Items

1. Further to my communication (Yeast Newsletter XXIX(2), p. 61-64) on microcalorimetric monitoring of yeast growth and metabolism, a literature reference list is now available, free of charge, upon request to LKB Produkter AB, RF Division, Box 305, S-161 26 Bromma, Sweden. Entitled "Literature Reference List: Bioactivity Monitor" the publication contains 50 references on yeast microcalorimetry and sections on viruses, bacteria, fungi, algae/protozoa, tissue cells, tissues/intact organs, invertebrates and ecology.

Brian Perry
present address:
B.S.G. Chelsea College
Hortensia Road
London, S.W. 10,
United Kingdom

2. Professor Ira Herskowitz has moved positions from the University of Oregon at Eugene to the University of California, Department of Biochemistry and Biophysics, San Francisco School of Medicine, San Francisco, California 94143, U.S.A.

3. In September 1982, I expect to have an opening for a postdoctoral fellow in my laboratory. The position is for one year, but can be renewed for a second and even a third year. The salary will be approximately $17,000. Interested candidates are invited to apply.

Enrico Cabib
Dept. of Health and Human Services
Bld. 10, Room 9N-115
National Institutes of Health
Bethesda, Maryland 20205 U.S.A.

4. Biotechnology - A comprehensive Treatise in 8 volumes. Editors are H.J. Rehm and G. Reed. Published by Verlag Chemie GmbH, Marketing Dept., P.O. Box 1260/1280, D-6940 Weinheim, West Germany. Subscription period for the Series (with considerable savings) ends June 30, 1982. Vol. I: Microbial fundamentals - to appear Dec. 1981.

Gerald Reed ed.
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