YEAST

A News Letter for Persons Interested in Yeast

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I. Microbiology Research Group of the South African Council for Scientific and Industrial Research. P. O. Box 395, Pretoria, South Africa.

Communicated by J. P. van der Walt.

Below follow the contributions from our laboratory:

Publications Which Have Appeared

- The yeast genus <u>Wickerhamiella</u> gen. nov. (Ascomycetes). J. P. van der Walt and N. v.d. W. Liebenberg; Antonie van Leeuwenhoek, Vol. 39 (1973), pp. 121-128.
- Candida homilentoma, a new yeast from South African insect sources.

 J. P. van der Walt and T. Nakase; Antonie van Leeuwenhoek, Vol. 39 (1973), pp. 449-453.
- Candida naeodendra, a new species of the Candida diddensii group.

 J. P. van der Walt, Elzbieta Johannsen and T. Nakase; Antonie van Leeuwenhoek, Vol. 39 (1973), pp. 491-495.
- Aessosporon dendrophilum sp. nov., the perfect state of <u>Bullera dendrophila</u>. J. P. van der Walt; Antonie van Leeuwenhoek, Vol. 39 (1973), pp. 455-460.
- Agglutinative mating types in <u>Saccharomyces transvaalensis</u>. J. P. van der Walt and N. v.d. W. Liebenberg; Antonie van Leeuwenhoek, Vol. 39 (1973), pp. 629-633.
- The perfect state of <u>Torulopsis magnoliae</u>. J. P. van der Walt and Elzbieta Johannsen; Antonie van Leeuwenhoek, Vol. 39 (1973), pp. 635-647.
- The dangeardien and its significance in the taxonomy of the ascomycetous yeasts. J. P. van der Walt and Elzbieta Johannsen; Antonie van Leeuwenhoek, Vol. 40 (1974), pp. 185-192.

Publications Appearing Shortly

- Ascospores in <u>Torulopsis</u> <u>dattila</u> (Kluyver) Lodder. J. P. van der Walt and Elzbieta Johannsen; Antonie van Leeuwenhoek, Vol. 40 Part 2 (1974).
- Ascorpores in the type of the genus <u>Torulopsis</u>. J. P. van der Walt and Elzbieta Johannsen; Antonie van Leeuwenhoek, Vol. 40 Part 2 (1974).
- Dr. Marie-Claire Pignal from the Départment Biologie Végétale of The Université Claude Bernard in Lyons spent four weeks in our laboratories. Apart from looking at yeasts, M1le Pignal also had the opportunity of observing the wild-life of the Kruger National Park.

II. Institute of Biochemistry and Physiology of Microorganisms. USSR Academy of Sciences, Pustchino Moscow region, USSR. Communicated by W. I. Golubev.

The following articles have recently been published:

Aksenov, S. I., Babjeva, I. P., Golubev, W. I., 1973. Mechanism of Microorganisms Adaptation to Extreme Low Humidity. "Life Science and Space Res. XI" (Proc. 15th Plenary Meeting of COSPAR. Madrid, Spain, May 21-24, 1972) Akad. Verlag-Berlin, 55-61.

Golubev, W. I., Vdovina, N. V., 1973. Yeast soil flora of rice fields treated by herbicides. "The behaviour, conversion and analysis of pesticides and their metabolites in soil" (The materials 1st All-Union Meeting, Pustchino-on-Oka, October 17-18, 1973), 66-73.

Kochetkov, N. K., Gorin, S. E., Sviridov, A. F., Chizhov, O. S., Golubev, W. I., Babjeva, I. P., Podelko, A. J., 1973. The polysaccharides of <u>Lipomyces</u> II. The Structure of Extracellular Glucuronomannan of <u>Lipomyces</u> <u>lipofer</u> 133. Izv. Akad. Nauk USSR (ser. chim.), N 10, 2304-2311.

Golubev, W. I., Babjeva, I. P., 1973. Taxonomic criteria of genus <u>Cryptococcus</u> Kütz. Mycologia i Phytopatologia, <u>7</u>, N6, 552-556.

Golubev, W. I., 1973. <u>Nadsonia commutata</u> nov. sp. Mikrobiologia, 42, N6, 1058-1061.

Babjeva, I. P., Golubev, W. I., Kartincev, A. V., Gorin, S. E., Zaslavskaja, P. L., 1973. The yeasts in the structure of forest and meadow biogenocenosises. Vest. MGU (biol., soil sci), N6, 67-73.

Golubev, W. I., Vdovina, N. V., 1974. The monosachharide composition of extracellular polysaccharides of some species of cryptococci. Mikrobiologia, 43, N1, 149-151.

The following article is in the press:

Golubev, W. I., Okunev, O. N., Vdovina, N. V., The assimilation of i-inositol by yeasts as the diagnostic sign. Mikrobiologia.

Works in progress:

The catabolism of i-inositol by cryptococci.

Electron microscopy of Nadsonia commutata.

III. United States Department of Agriculture. Northern Regional Research Laboratory. Peoria, Illinois 61604. Communicated by C. P. Kurtzman.

The following is a summary of an article in press in <u>Mycopathologia</u> et <u>Mycologia Applicata</u>.

SCANNING ELECTRON MICROSCOPY OF ASCOSPORES OF DEBARYOMYCES

AND SACCHAROMYCES

Ъy

C. P. Kurtzman, M. J. Smiley and F. L. Baker

Abstract

Ascospores from species of Debaryomyces and the Torulasporagroup of Saccharomyces were examined by scanning electron microscopy. Ornamentation on ascospores of D. hansenii varied from short to long interconnected ridges or broad based, elongated, conical protuberances. A spiral ridge system was detected on the ascospores of D. marama, but wart-like protuberances occurred on those of D. cantarellii, D. castellii, D. coudertii, D. formicarious, D. phaffii, D. vanriji and D. yarrowii. Ascospores of D. halotolerans did not have protuberances and the species appears to be identical with Pichia farinosa. Wart-like protuberances also were found on ascospores of S. delbrueckii, S. microellipsodes, S. rosei, S. inconspicuus, S. fermentati, S. montanus and S. vafer, but the ascospore surface of S. pretoriensis was covered by fine ridges. Short tapered ridges covered the ascospores of S. kloeckerianus.

IV. Department of Biology, McMaster University, Hamilton, Ontario, Canada.

Communicated by J. J. Miller.

We have recently compared the fine structure of nonsporulated cells and asci of \underline{S} . cerevisiae with the freeze-etching technique.

Nonsporulated cells differed markedly from vegetative cells and tended to resemble asci or ascospores. Unlike vegetative cells, few of them had a single central vacuole, but exhibited a number of small vaculoes containing striated, amorphous or granular material; their walls were thicker than those of vegetative cells and possessed an outer fibrillar and an inner particulate zone, features found characteristic of the ascus walls.

These observations indicate that although certain cells of a yeast population do not produce ascospores in acetate sporulation medium, they nevertheless may undergo some of the structural changes characteristic of sporulating cells. In this connection it is of interest that nonsporulated cells survived much longer under desiccation than vegetative cells and they also, like spores, required much more time than vegetative cells to produce buds when transferred to germination medium.

Recent Publication

Steele, S. D. and J. J. Miller. Pseudomycelial development and sporulation in <u>Saccharomyces fragilis</u>. Can. J. Microbiology 20: 265-267. 1974.

In Press

- Steele, S. D. and J. J. Miller. Ultrastructural changes in germinating ascospores of <u>Saccharomyces</u> <u>cerevisiae</u>. Can. J. Microbiology.
- Steele, S. D. and J. J. Miller. Similarities between non-sporulated yeast cells and asci. Can. J. Microbiology.
- V. <u>Departamento de Microbiología, CSIC y Facultad de Ciencias, Universidad de Salamanca.</u> Salamanca, Spain. <u>Communicated by J. R. Villanueva.</u>

The articles below have appeared recently.

- Villanueva, J. R. and Gacto, M. (1973) "Characterization of β -(1+3) glucanases of yeast". Proc. Third. Internat. Spec. Symp. on Yeast. Part II. pp. 261-283.
- Duran, A., Uruburu, F. and Villanueva, J. R. (1973) "Morphogenetic and nutritional studies of Geotrichum lactis cells". Arch. Mikrobiol. 88, 245-256.
- Nombela, C., Uruburu, F. and Villanueva, J. R. (1974) "Studies on membranes isolated from extracts of <u>Fusarium culmorum</u>". J. Gen. Microbiol. 81, pp. 247-254.
- Sierra, J. M. Sentandreu, R. and Villanueva, J. R. (1973) "Regulation of wall synthesis during <u>Saccharomyces</u> cerevisiae cell cycle". FEBS Letters <u>34</u>, pp. 285-290.
- Martín, J. F., Nicolas, G. and Villanueva, J. R. (1973) "Chemical changes in the cell walls of conidia of <u>Penicillium notatum</u> during germination". Can. Microbiol. 19, 789-796.
- Martín, J. F., Uruburu, F. and Villanueva, J. R. (1973) "Ultrastructural changes in the conidia of <u>Penicillium notatum</u> during germination". Can. J. Microbiol. 19, pp. 797-801.
- García-López, M. D., Laborda, F., Uruburu, F. and Villanueva, J. R. (1973) "Identification of a lytic microorganism isolated from the soil as a strain of <u>Streptomyces flavovirens</u>". Japan. J. Microbiol. 17, 223-227.
- Villanueva, J. R., Gacto, M. and Sierra, J. M. (1974) "Enzymatic composition of a lytic system from Micromonospora chalcea AS" in Yeast, Mould and Plant Protoplasts (Ed. Villanueva, J. R., García Acha, I., Gascón, S. and Urburu, F.) Academic press. pp. 3-25.

- Gascón, S., Lazo, P. S., Moreno, F. and Ochoa, A. G. (1974) "Secretion of invertase and α-galactosidase by yeast" In: Yeast, Mould and Plant Protoplasts. Academic Press pp. 157-167.
- Sentandreu, R. and Elorza, M. V. (1974) "The biosynthetic pathway of yeast mannan glycoproteins". In: Yeast, Mould and Plant Protoplasts. Academic Press pp. 187-204.
- Elorza, M. V. and Sentandreu, R. (1974) "The effect of cycloheximide on mannosyl transferase activity of a membrane preparation from Saccharomyces cerevisiae" In: Yeast, Mould and Plant Protoplasts. Academic Press pp. 205-210.
- VI. Department of Food Science and Technology, University of California, Davis, Ca. 95616. Communicated by H. J. Phaff.

Below follow the abstracts of recent work:

1. Lysis of Yeast Cell Walls: Glucanases from Bacillus circulans WL-12

Graham H. Fleet and Herman J. Phaff
Journal of Bacteriology, in press (July issue 1974)
Abstract

Endo- β -(1+3)- and endo- β -(1+6)-glucanase are produced in high concentration in the culture fluid of Bacillus circulans WL-12 when grown in a mineral medium with baker's yeast cell walls as the sole carbon source. Much lower enzyme levels were found when laminarin, pustulan or mannitol were the substrates. The two enzyme activities were well separated during Sephadex G-100 chromatography. The endo- β -(1+3)-glucanase was further purified by DEAE-cellulose and hydroxyapatite chromatography, while the endo- β -(1-6)-glucanase could be purified further by passage over DEAEcellulose and carboxymethylcellulose chromatography. The endo-β- $(1\rightarrow 3)$ -glucanase was specific for the β - $(1\rightarrow 3)$ -glucosidic bond but it did not hydrolyze laminaribiose; laminaritriose was split very slowly. β -(1-4)-Bonds in oat glucan in which the glucosyl moiety is substituted in the 3-position were also cleaved. The kinetics of laminarin hydrolyses (optimum pH 5.0) were complex but appeared to follow Michaelis-Menten theory, especially at the lower substrate concentrations. Glucono- δ -lactone was a non-competitive inhibitor and ${\rm Hg}^{Z+}$ inhibited strongly. The enzyme has no metal ion requirements or essential sulfhydryl groups. The purified β -(1 \rightarrow 6)-glucanase has an optimum pH of 5.5 and its properties were studied in less detail. In contrast to the crude culture fluid the two purified β-glucanases have only a very limited hydrolytic action on cell walls of either baker's yeast or of Schizosaccharomyces pombe. Although our previous work had assumed that the two glucanases studied here are responsible for cell wall lysis, it now appears that the culture fluid contains in addition a specific lytic enzyme which is eliminated during the extensive purification process.

Demonstration of a Fibrillar Component

In the Cell Wall of the Yeast

Saccharomyces cerevisiae and its chemical nature

Marie Kopecká (Department of General Biology, Faculty of Medicine, J. E. Purkyne, University Brno, Czechoslovakia), H. J. Phaff, and G. H. Fleet.

The Jour. Cell Biology. In press (July issue 1974)
Abstract

The ultrastructure of isolated cell walls of Saccharomyces cerevisiae from the log and stationary phases of growth was studied after treatment with the following enzymes: purified endo- β -(1 \rightarrow 3)-glucanase and endo- β -(1 \rightarrow 6)-glucanase produced by Bacillus circulans; purified exo- β -glucanase and endo- β -(1+3)glucanase produced by Schizosaccharomyces versatilis; commercial Pronase. While exo-β-glucanase from S. versatilis had no electronmicroscopically detectable effect on the walls, Pronase removed part of the external amorphous wall material disclosing an amorphous wall layer in which fibrils were indistinctly visible. Amorphous wall material was completely removed by the effect of either endo- β -(1+3)- or endo- β -(1+6)glucanase of B. circulans or by a mixture of the two enzymes. As a result of these treatments a continuous fibrillar component appeared, composed of densely interwoven microfibrils resisting further action by both of the B. circulans enzymes. The fibrillar wall component was also demonstrated in untreated cell walls by electron microscopy after negative staining. Because of the complete disappearance of the fibrils following treatment with the S. versatilis endo- β -(1+3)-glucanase it can be concluded that this fibrillar component is composed of β -(1+3)-linked glucan. Bud scars were the only wall struutures resistent to the effect of the latter enzyme.

3. <u>Cryptococcus cereanus</u> a new species of the genus <u>Cryptococcus</u>

H. J. Phaff, M. W. Miller and Mary Miranda

Department of Food Science and Technology, University of California, Davis, Ca. 95616; W. B. Heed, and W. T. Starmer, University of Arizona, Tucson, Ar 85721.

Submitted to: Intern. Jour. Syst. Bacteriol.

Abstract

A novel representative of the yeast genus <u>Cryptococcus</u> has been recovered eleven times during 1971, 1972 and 1973 from soft-rot pockets in three species of cactus, <u>Cereus schottii</u> Engelm., <u>Cereus giganteus</u> Engelm., and <u>Cereus thurberi</u> Engelm. The collections were made in the Sonoran desert of Southern Arizona and of Northern Mexico. The rot pockets constitute important breeding sited for <u>Drosophila</u> spp. which presumably feed on the yeasts present. The new species was named after the genus of cactus with which it is associated.

4. The following is an abstract of a paper presented at the annual meeting of the Am. Soc. for Microbiology, Chicago, May 1974.

Deoxyribonucleic Acid (DNA) Base Composition and DNA-DNA Hybridization Studies among Species of the Yeast Genus Schwanniomyces. C. W. Price and H. J. Phaff, University of California, Davis, California.

DNA was extracted from the type strains of S. occidentalis, S. alluvius, S. castellii, and S. persoonii. An additional strain of S. persoonii exhibiting unusual ascospore morphology was also studied. Mole percent guanine and cytosine (GC%) of the nuclear DNA was calculated from buoyant density values determined by isopycnic density gradient centrifugation. extent of DNA-DNA hybridization was assayed by both the nitrocellulose filter method of Denhardt and by the hydroxy apatite batch technique of Brenner. DNA of all of the strains of Schwanniomyces had a mean GC% of 35.1% and a range of 35.0-35.2. DNA from the type species S. occidentalis exhibited a high degree of homology with DNA from all of the other Schwanniomyces strains, and little homology with DNA from yeasts with similar life cycles: Debaryomyces cantarellii (GC% = 35%), Saccharomyces rosei (GC% = 43%), and Pichia vini (GC% = 39%). We propose that the presently accepted four species of Schwanniomyces be reduced to a single species, Schwanniomyces occidentalis, and that the other three species be considered as varieties, based on sugar utilization reactions. It may be that the strain with unusual spore morphology represents a natural mutant forming crater-like depressions rather than protuberances on the spore wall.

- 5. Dr. Carlos Hardisson, University of Oviedo, Spain, has arrived in Davis on June 6 to spend three months in our laboratory. He will participate in several research projects.
- 6. The following paper has been published: DNA Base Composition and Hybridization Studies on the Human Pathogen Sporothrix schenkii and Ceratocystis species. Leda C. Mendonca-Hagler, L. R. Travassos, K. O. Lloyd and H. J. Phaff. Infection and Immunity 9, 934-938, 1974.
- VII. Fimbriae, fusion and flocculation in yeasts. A. W. Day and N. H. Poon,

 Department of Plant Sciences, University of Western Ontario, London,

 Canada.

We have recently discovered that the cells of several species of yeasts and yeast-like fungi produce large numbers of fine hairs on the cell wall. These hairs have been termed fimbriae, because of their close developmental & morphological similarity to bacterial <u>fimbriae</u> (or <u>pili</u>).

The fimbriae of the basidiomycete <u>Ustilago violacea</u> have been studied in most detail. These fimbriae are about 60-70 A° wide, and vary in length to over 10 μm (i.e. longer than the cell itself- about 6 μm). They appear to be composed mainly and perhaps wholly of protein. When cells are defimbriated by high speed agitation new fimbriae regenerate in about one hour. This regeneration is prevented by

cycloheximide and rifampin but not by chloramphenical, and therefore appears to depend on \underline{de} novo protein synthesis on cytoplasmic ribosomes.

The fimbriae of <u>U. violacea</u> cannot regenerate at temperatures above 26°C and neither can the cells conjugate at this temperature. However, they do grow well at 26-28°C. This and other evidence strongly suggests that the fimbriae are necessary for conjugation. Our current evidence suggests that they may act as conduits between the conjugating cells, permitting exchange of macromolecules at a time when the cells are in close physical contact, but with intact cell walls. They may also play a role in establishing the close contact i. e., in cellular agglutination.

Similar long fimbriae have been observed in <u>Ustilago maydis</u> and <u>Leucosporidium (Candida) scottii</u>. However in <u>Saccharomyces cerevisiae</u>, <u>Hansenula wingei</u>, <u>H. saturnus</u>, <u>Schizosaccharomyces pombe</u>, <u>S. octosporus</u>, <u>Torulopsis utilis</u>, <u>Nadsonia sp., Rhodosporidium sp. (Rhodotorula glutinis</u>), and <u>Lipomyces lipofer</u>, we find very short fimbriae (0.5 μ m long) densely covering the cells. None of the species we have examined so far has been devoid of fimbriae. These short fimbriae can be seen in water washed metal shadowed preparations, but we have been able to display them much better after washing the cells with ether.

These results indicate that fimbriae may play a major role in flocculation. Haploid cells of either mating-type also produce sparse short fimbriae some of which produce a terminal bulb. It seems likely that these fimbriae are involved in conjugation in Saccharomyces, in view of the results we have obtained with \underline{U} . $\underline{violacea}$.

A detailed description of these results will be published shortly.

VIII. National Research Council Canada. Division of Biological Sciences.
Ottawa, Canada KlA OR6. Communicated by Byron Johnson.

CELL DIVISION IN YEASTS. II. TEMPLATE CONTROL OF CELL PLATE BIOGENESIS IN SCHIZOSACCHAROMYCES POMBE

Byron F. Johnson, Division of Biological Sciences, National Research Council of Canada, Ottawa, Ontario KlA OR6

Bong Y. Yoo, Department of Biology, University of New Brunswick,

and

G. C. Calleja, Division of Biological Sciences, National Research Council of Canada, Ottawa, Ontario KiA OR6

SUMMARY

Fluorescence micrographs of fission yeast cells in cell division show a bright ring which grows centripetally to finally become a bright disk bisecting the cell. The behaviour of the brightly fluorescent structure corresponds in detail with that of an electron transparent structure previously described in the cell plate. The electron transparent structure acts as a template, upon which the new ends of the daughter cells are formed. At fission the brightly fluorescent template is removed concomitant with release of the two daughter cells. The stepwise sequence of generation and removal should allow closer scrutiny of cell division controls in yeasts than has heretofore been possible.

In Press: Cell Cycle Controls, Academic Press (late 1974); Eds: G. M.
Padilla, I. L. Cameron and A. M. Zimmermann

On the Nature of the Forces Involved in the Sex-directed Flocculation of a Fission Yeast

G. B. Calleja

Division of Biological Sciences
National Research Council of Canada
Ottawa, Ontario K1A OR6

In Press: Can. J. Microbiol.

SUMMARY

Cells of Schizosaccharomyces pombe NCYC 132 flocculate during stationary growth phase. Inducible by aeration, flocculation is followed by conjugation and formation of ascospores. Not all cells in a culture are competent to form flocs. Cells in flocs are separable from the incompetent cells by differential sedimentation. Stable when diluted in de-ionized water, flocs are reversibly deflocculated by heat. T_, the temperature at which half a floc population is deflocculated, is 61°C in deionized water. Mono- and divalent cations increase T_ and the rate of reflocculation at ambient temperature. Guanidinium chloride, urea, sodium dodecyl sulfate, extremes of pH, and ultrasonication also reversibly deflocculate purified flocs. In contrast, proteinases, e.g., pronase and trypsin, cause irreversible deflocculation. All reflocculation exhibits hysteresis. It is very likely that the principal binding forces are hydrogen and hydrophobic bonds between walls of competent cells, rather than ionic bonds. The data also suggest the involvement of some proteinaceous structure on the cell wall.

L. Zeman and C. V. Lusena Closed circular DNA associated with yeast mitochondria. FEBS Letters (1974) 38, 171-174.

The $\underline{\text{in}}$ $\underline{\text{vivo}}$ labelling of the 2μ DNA circles of nuclear density is inhibited by cycloheximide in analogy with the behaviour of the nuclear and not the mitochondrial DNA. Contrary to some reports our circular DNA fraction also contains DNA of mitochondrial density.

L. Zeman and C. V. Lusena
DNA precursors and the absence of thymidine kinase in yeast
 mitochondria.
FEBS Letters (1974) 38, accepted for publication.

Yeast mitochondria do not contain any detectable levels of thymidine kinase and among thymine derivatives tested only dTDP and dTTP could be used as effective precursors for the $\underline{\text{in}}$ $\underline{\text{vitro}}$ labelling of m-DNA.

IX. <u>Karl-Marx University</u>, <u>Klinik für Hautkrankheiten</u>, <u>Liebigstr. 21</u>. 701 <u>Leipzig</u>, <u>DDR</u>. <u>Communicated by Christina Schönborn</u>.

We have determined the sodium chloride tolerance of about 200 freshly isolated yeast strains from the human microflora. The salt concentration in the nutrient medium (glucose-peptone-agar) was the limiting factor for growth in this medium, and the various species showeddifferent sensitivities to increasing NaCl concentrations. Growth-limiting NaCl values below 8% were observed for strains of Candida krusei, Candida pseudotropicalis, and Cryptococcus neoformans. A maximal tolerans for NaCl (14.8%) was observed for Candida parapsilosis and C. guilliermondii. Average values are given in the following table:

Species	Number of Strains	Limiting NaCl concentrations for growth
Cryptococcus neoformans	2	5.4
Candida krusei	16	8.4
Candida pseudotropicalis	16	8.8
Candida humicola	9	9.7
Trichosporon cutaneum	17	10.1
Rhodotorula mucilaginosa	12	10.3
Torulopsis glabrata	9	10.8
Candida albicans	20	10.9
Candida tropicalis	20	11.7
Candida lipolytica	12	11.9
Candida guilliermondii	20	13.7
Candida parapsilosis	20	14.2

X. <u>Mikrobiologisches Institut</u>, <u>Swiss Federal Institute of Technology</u>, <u>Weinbergstrasse 38</u>, <u>8006 Zürich</u>, <u>Switzerland</u>. <u>Communicated by A.</u> Fiechter.

Recent Publications:

- H. W. Blanch and I. J. Dunn: Modelling and Simulation in Biochemical Engineering. Adv. Biochem. Eng. 3, 127-165 (1974).
- H. W. Blanch and A. Einsele: The Kinetics of Yeast Growth on Pure Hydrocarbons. Biotechnol. Bioeng. 15, 861-877 (1973).
- S. Divjak and J. -R.Mor: On the Activity of Carbon Dioxide Fixation in growing Yeasts. Arch. Microbiol. 94, 191-199 (1973).
- U. Flury, B. Heer, and A. Fiechter: Isoenzyme Pattern of Malate Dehydrogenase during Respiratory Derepression in <u>Schizo-saccharomyces pombe</u>. Arch. Microbiol. <u>97</u>, 141-148 (1974).
- U. Flury, B. Heer, and A. Fiechter: Regulatory and Physico-Chemical Properties of two Isoenzymes of Malate Dehydrogenase from <u>Schizosaccharomyces</u> pombe. Biochim. Biophys. Acta. 341, 465 (1974).
- K. E. Weibel, J. -R.Mor, and A. Fiechter: Rapid Sampling of Yeast Cells and Automated Assays of Adenylate, Citrate, Pyruvate and Glucose-6-Phosphate Pools. Anal. Biochem. 58, 208-216 (1974).

In Press:

- H. Schatzmann and A. Fiechter: Methodik des anaeroben Chemostaten. Pathol. Microbiol. (1974).
- XI. National Aeronautics and Space Administration, Ames Research Center, Moffett Field, California 94035. Communicated by T. Satyanarayana.

The following is a summary of an article accepted for publication in Biochim. Biophys. Acta (1974).

EVIDENCE FOR TWO IMMUNOLOGICALLY DISTINCT ACETYL-COENZYME A SYNTHETASES IN YEAST

bу

T. Satyanarayana, Adrian D. Mandel and Harold P. Klein

SUMMARY

1. In this investigation, some immunological properties of the yeast acetyl-CoA synthetase (acetate: CoA ligase (AMP), EC 6.2.1.1) elaborated under aerobic and non-aerobic conditions are presented.

- 2. The antibody produced by each enzyme is immunologically specific. No evidence was found for the presence of inhibitory agent(s) in either enzyme extract which inhibited the heterologous enzyme antibody reaction.
- 3. Aerobic crystalline enzyme activity was inhibited 85% by its homologous antiserum which inhibited non-aerobic activity by only 20%, while non-aerobic enzyme activity was inhibited 65% by its homologous antiserum, which inhibited aerobic activity by about 13%.
- 4. Data presented in this paper indicate the presence of two distinct acetyl-CoA synthetases in this strain of yeast.
- XII. <u>University of Tokyo, College of General Education</u>, <u>Institute of Physics</u>, <u>Komaba 3-8-1</u>, <u>Tokyo 153</u>, <u>Japan</u>. <u>Communicated by Takashi Ito</u>.

The following are the summaries of recent papers now in the process of publication.

(1) $\underline{\text{In}}$ $\underline{\text{vivo}}$ Evidence for the Participation of Singlet Excited Oxygen Molecules in the Photodynamic Inactivation.

Takashi Ito and Katsumi Kobayashi

Sci. Pap. Coll. Gen. Educ. Univ. Tokyo 24, 33-36 (1974).

SUMMARY

- D₂0, known to enhance the decay lifetime of singlet oxygen, and azide ion, known as a specific quencher of singlet oxygen, were applied to a cell system to see how they affect the photodynamic inactivation. The results showed that the acridine orange (AO)-sensitized photodynamic inactivation of yeast cells was enhanced in going from normal to perdeuterated medium, while it was markedly reduced (protected) in the presence of azide ions in aqueous suspension. This set of specific pertubations is, therefore, taken to mean that singlet excited oxygen molecules are participating as the intermediate species in in vivo photodynamic action.
 - (2) Uptake and Localization of Acridine Orange in Yeast Cells
 Takashi Ito

Sci. Pap. Coll. Gen. Educ. Univ. Tokyo 24, 37-44 (1974).

SUMMARY

In a few yeast strains, the uptake of acridine orange (A0) was determined by means of spectrophotometry and radioactivity measurements of bound ³H-A0 for stationary phase and log phase cultures under different staining conditions. The uptake of log phase culture was considerably higher than that of stationary culture. The amount of uptake was also dependent on the ploidy. By parallel observations of the intracellular fluorescence

patterns of such cells, it was found that in the stationary phase cell much of the AO molecules were localized in a small spot and that in the log phase cell they were accumulated over the entire cell space. All of these cells were alive in the dark when tested on agar plates. In view of the specific interaction of acridine dyes with nucleic acid components in the cell, these results will be useful for undertaking further experiments leading to the elucidation of dye-mediated photoeffects on yeast cells.

XIII. Alko, Box 350, 00101 Helsinki 10, Finland. Communicated by Heikki Suomalainen.

THE USE OF A BIOCHEMICAL EQUATION AS A MODEL FOR YEAST GROWTH

by E. Oura

3 Symp. Tech. Mikrobiol., Berlin 1973, Institut für Gärungsgewerbe und Biotechnologie, Berlin 1973, pp. 355-360, Kurzreferate 48H (in German).

A reaction equation was developed to describe the growth of yeast on glucose. Special attention was given to the participation of adenosine and respiratory mucleotides in the growth process. The following theoretic values, e.g., can be estimated from this kind of equation: energy relations in cultivation with different substrates or growth conditions; the ATP-turnover number; the amount of glycerol formed or CO₂ assimilated. Further, it is possible to estimate the actual rate of enzymic reactions in vivo during growth under various conditions.

α-GLUCOSIDASE ACTIVITY AND LEAVENING CAPACITY OF BAKER'S YEAST

by H. Suomalainen and E. Oura

3 Symp. Tech. Mikrobiol., Berlin 1973, Institut für Gärungsgewerbe and Biotechnologie, Berlin 1973, pp. 361-366 (in German)

No correlation has been found between the α -glucosidase activity and the leavening capacity of different brands of baker's yeast. Glucose, fructose, sucrose and raffinose were used first during the leavening of dough, maltose was fermented only after the hexose content had decreased to a low level. The decrease in sugar content of the leavening dough correlated well with the amount of CO_ obtained during leavening. The leavening capacity was not influenced by the addition of either saccharase or β -amylase, but glucoamylase clearly improved the results of proofing.

SOME ENZYMOLOGICAL FACTORS INFLUENCING THE LEAVENING CAPACITY AND KEEPING QUALITY OF BEAKER'S YEAST

by H. Suomalainen
Paper presented at the 8th International ICC Working and
Discussion Meetings, May 16-18, 1974, Vienna, Austria.

The paper deals with α -glucosidase and the leavening capacity of baker's yeast, changes in commercial baker's yeast during its ripening period and the effect of storage on the nucleic acid composition of baker's yeast. During the ripening period yeast cells reach a uniform single cell stage, which has better storage qualities than budding cells. Also the synthesis of messenger-RNA and the other RNA species was reduced, leading to a reduced rate of protein synthesis. The other results have been presented in detail earlier.

PARTIAL PURIFICATION AND CHARACTERIZATION OF A 3 5 5 CYCLIC AMP

PHOSPHODIESTERASE FROM SACCHAROMYCES CEREVISIAE by J.L. Londesborough

Paper presented at the 545th Meeting of the Biochemical Society, December 1973, Swansea, England. The paper will be published in Biochemical Society Transactions.

The diesterase was purified 2800-fold from the 34 000 g supernatant of mechanically disintegrated yeast by treatment with hydroxyapatite, gel-filtration on Sephadex G-100, and TEAE-cellulose chromatography. 5° AMP was the only product of the hydrolysis of cyclic AMP. The enzyme had a molecular weight of 70 000 and a pI of 5.1. The pH optimum was 8.0. Importantly, the enzyme does not require added divalent metal ions, and is not inhibited by EDTA. The K was 50 μM . Theophylline is a competitive inhibitor. Thiols, such as mercaptoethanol, cysteine etc., also caused large inhibitions.

PEAT AS SUBSTRATE FOR YEAST CULTIVATION

by E. Oura and K. Silla

Abstract of paper to be presented at 15de Nordiska Kemistmötet, June 7-11, 1974, Tampere, Finland. The abstract will be published in Resumeer (in Swedish).

The slightly humified peat containing about 40-50% of cellulose and hemicellulose was hydrolysed for two hours in 0.3-1.0% sulphuric acid at 125°C. More than 60% of the peat remained as a remnant and the peat extract contained only about 35% of peat dry matter. Half of it was reducing sugars, mainly glucose, galactose and xylose. The peat extract was concentrated to half its volume and Candida utilis was grown in it. The yield in aerobic growth was about 10-12 g yeast d.m./100 g dry peat. We have also followed the ethanol formation when Saccharomyces cerevisiae was grown anaerobically in acid hydrolysate. The final alcohol content in the medium was 0.5% (w/v). The peat was also hydrolysed with enzymes (cellulase-hemicellulase) in a system where the peat and the enzymes were separated from the growth suspension with a permeable membrane. In this case the yield was 15-20 g yeast d.m./100 g dry peat.

PHOSPHOENOLPYRUVATE CARBOXYKINASE ACTIVITY AND ITS DEPENDENCE ON CATABOLITE REPRESSION IN BAKER'S YEAST

by S. Haarasilta and E. Oura

Abstract of paper to be presented at 15de Nordiska Kemistmötet, June 7-11, 1974, Tampere, Finland. The abstract will be published in Resumeer (in Swedish).

In aerobic batch cultivations on glucose, where the growth of yeast is to be considered as glucose-ethanol biphasic growth, PEPCK activity appeared only after glucose was consumed. Further, under anaerobic continuous cultures with glucose as the carbon source of yeast, the synthesis of PEPCK was fully blocked and under aerobic conditions only negligible activity of PEPCK was observed. Galactose, like glucose, repressed the synthesis of PEPCK. The presence of aspartic acid affected neither the activity of PEPCK nor the time of the appearance of activity during the biphasic growth. Significant amounts of PEPCK could be observed only in yeast grown on pyruvate or on ethanol, also under conditions free from catabolite repression. Besides the catabolite repression, glucose regulated PEPCK via another mechanism. When glucose was added to the medium of yeast growing on ethanol, the PEPCK activity of yeast dropped by more than 50% in 30 min. This phenomenon is to be considered as a glucose induced inactivation of PEPCK.

LOCALIZATION OF PYRUVATE CARBOXYLASE AND PHOSPHOENOLOPRUVATE CARBOXYKINASE IN BAKER'S YEAST.

by L. Taskinen and S. Haarasilta

Abstract of paper to be presented at 15de Nordiska Kemistmötet, June 7-11, 1974, Tampere, Finland. The abstract will be published in Resumeer (in Swedish).

The localization of pyruvate carboxylase (PC) and phosphoenol-pyruvate carboxykinase (PEPCK) in baker's yeast grown on glucose or ethanol has been investigated by centrifugation of yeast homogenates in continuous saccharose gradients. The density distribution of PC, PEPCK, and marker enzymes was determined, and fractions further identified by electron micrography. When mechanically disintegrated yeast preparations were used, most of the PC activity was detected in fractions where the density was under 1.05, i.e., in the region of the soluble enzymes. Only negligible amounts of the PC activity could be detected in the microsomal (1.05-1.15) and mitochondrial (1.15-1.20) density ranges. PEPCK activity could be detected only in the density range of the soluble enzymes. This shows, that PEPCK is an extramitochondrial enzyme. The distribution of PC activity was independent of whether yeast was grown on glucose or ethanol.

Although most of the PC activity and all of the PEPCK activity in the previous experiments was detected in the region of the soluble enzymes, this does not necessarily indicate, that these enzymes are cytosolar. In preliminary experiments, where spheroplast preparations, lysed by mild osmotic shock, were used in the fractionation, we could clearly observe PC activity in the microsomal region.

FRACTIONATION AND ELECTRON MICROSCOPY OF CELL ENVELOPES FROM BAKER'S YEAST

by L. Taskinen

Abstract of paper to be presented at the Annual Meeting of the Scandinavian Society for Electron Microscopy (SCANDEM-74), June 10-12, 1974, Helsinki, Finland. Submitted for publication in J. Ultrastruct. Res.

Washed cell envelopes of aerobic and anaerobic baker's yeast (Saccharomyces cerevisiae) obtained by differential centrifugation were disrupted mechanically. Further fractionation of the envelope homogenate containing fragments of cell wall and plasma membrane was made by swing-out or zonal centrifugations in stepwise or continuous sucrose density gradients. Evidence for the purity and intact nature of plasma membrane was got by measuring the distribution pattern of the activity of marker enzymes and by electron micrography. Selected particulate fractions were prefixed with 3% glutaraldehyde buffered with 0.2 M phosphate (pH 7.2) for 2 hours at 4°C. After fixation with chilled 1 per cent osmium tetroxide for 2 hours preparations were subjected to dehydration with increasing series of ethanol and propylene oxide before Epon embedding. Thin sections were poststained with 3 per cent uranyl acetate and 1 percent lead citrate.

Although there are as yet no adequate biochemical criteria for the purity of yeast plasma membranes, the results suggest that the present, simple, method of preparation is more reliable than others currently available.

THE EFFECT OF THE INTENSITY OF AERATION ON THE BIOCHEMICAL COMPOSITION OF BAKER'S YEAST. I. FACTORS AFFECTING THE TYPE OF METABOLISM.

11. ACTIVITIES OF THE OXIDATIVE ENZYMES.

by E. Oura

Accepted for publication in Biotechnol. Bioeng.

The abstract of part I:

A cultivation method is presented which permits the study of the effects of aeration intensity under conditions where the influence of catabolite repression is eliminated. A completely synthetic medium with glucose as the only carbon and energy source is also described. The capacity of yeast to perform aerobic varies when cultivated under different intensities of aeration. A clear maximum is observed at growth with 10% oxygen in the aerating gas mixture. In conditions where catabolite re-

pression does not function yeast has the potential for oxidative metabolism even under oxygen-limited growth. The main agent controlling the ability of yeast to support growth using solely oxidative metabolism is the available oxygen. At high oxygen tensions the metabolism is disturbed.

The abstract of part II:

When the effect of catabolite repression is eliminated Saccha romyces cerevisiae prefers an aerobic metabolism. The potential for completely aerobic catabolism exists in circumstances where its action is limited by the oxygen available. When the oxygen absorption in the medium is adequate, yeast uses a solely oxidative metabolism for energy-yielding reactions. The changes observed in the activity of malate dehydrogenase can be described as a function of two isoenzymes, both of which are affected by oxygen; the isoenzyme participating in the glyoxylate cycle shows variations in activity similar to that observed in isocitrate lyase. NAD-linked glutamate dehydrogenase activity roughly follows that of malate dehydrogenase and isocitrate lyase; in cultivations with the same growth rate the NADP-linked dehydrogenase is insensitive to the oxygen level. The cytochromes aa,, b and c have a clear maximum at low oxygen tension, the most sensitive being cytochrome aa,. The imbalance between cytochrome c:oxygen oxidoreductase activity and the amount of cytochrome aa, and the correlation observed between respiration rate and the activity of NADH,: cytochrome c reductase are discussed.

ON THE ENVELOPE GLYCOLIPIDS OF BAKER'S YEAST

by K. Työrinoja, T. Nurimen and H. Suomalainen

Biochem. J. (in press)

Sphingolipids were found to dominate in the glycolipids from the cell envelope of baker's yeast. A relatively large quantity of ceramides was detected. Among the several complex phosphosphingolipids described, ceramide-(P-inositol) -mannose was the main component. About 55% of long-chain bases in sphingolipids consisted of C_{18} -phytosphingosine. Other bases, found in decreasing concentrations, were C_{20} -phytosphingosine, C_{20} -dihydrophytosphingosine, C_{18} -dihydrosphingosine and C_{19} -dihydrosphingosine. The presence of sterolglycosides, sulfolipids, cerebrosides and acyglycoses was demonstrated.

XIV. <u>Institut für Physiologische Chemie der Ruhr-Universitat Bochum.</u>
<u>Communicated by W. Duntze.</u>

The following is an abstract of a recent article.

Purification and Partial Characterization of α -Factor, a Mating-Type Specific Inhibitor of Cell Reproduction from Saccharomyces cerevisiae

Wolfgang Duntze, Dieter Stötzler, Elizabeth Bücking-Throm, and Sigrid Kalbitzer

Cells of Saccharomyces cerevisiae exhibiting the α mating type excrete into the culture medium a low-molecular-weight substance, termed α factor. This factor, which specifically inhibits DNA replication in cells of the opposite mating type a, has been purified more than 100,000-fold from culture filtrates of α cells. Purified α factor appears to be homogeneous as judged from thin-layer chromatography and thin-layer electrophoresis in different systems. It shows a positive nihydrin reaction and, upon hydrolysis, gives rise to several ninhydrin-positive substances of which the amino acids leucine, glycine, proline, glutamic acid, tyrosine, trytophan and possibly histidine have been identified. The presence of tryptophan and tyrosine is also confirmed by the ultraviolet absorption spectrum of the factor. In addition, purified α factor contains cupric ions which can be separated from the ninhydrin-positive material by thinlayer electrophoresis at pH 3.6. Gel filtration on Sephadex G-25 in 8 M urea indicates a molecular weight in the range of 1400. The properties of the purified α factor are consistent with those of a low-molecular-weight peptide.

XV. <u>Laboratoire des Fermentations de l'Institut Pasteur, 75 Paris Xvè, Station de Technologie des Produits végétaux, INRA, 21 Dijon (France).</u>
Communicated by P. Bréchot.

ACTION ON OLEANOLIC ACID
ON GLUTAMATE DEHYDROGENASE ACTIVITY

Proceedings of the Third International Specialized Symposium on Yeasts, Otaniemi, Finland, 1973. Part 1, Abstracts pp. 130-131.

P. BRECHOT, P. DUPUY, Mme M. CROSON

We have already shown that oleanolic acid acts as a yeast anaerobic growth factor (1). In order to explain the physiological effect of this substance the influence of this triterpenic acid on the activity of glutamate dehydrogenase (GDH) has been studies: this enzyme seems to play a basic part in protein synthesis, (2) moreover, Tomkins et al. have pointed out the blocking effect of diethylstilbestrol (DES), a synthetic steroid oestrogen, on bovine GDH activity. (3).

In our experiments bovine (4) and yeast (5) GDH have been used under conditions to observe comparative results with both enzymes. Enzyme activities have been measured by observance at 340 nm using the specific coenzyme for each enzyme, NAD for bovine GDH, NADP for yeast GDH.

Our results show that oleanolic acid has no action on the activity of either bovine or yeast GDH, when the reductive amination of ketoglutarate is studied.

With the opposite reaction-oxidative deamination of glutamateoleanolic acid has an inhibitive effect on the activity of yeast GDH, 10 to 20%; with bovine GDH the inhibition is more important: 50%.

Consequently oleanolic acid exhibits a limited but actual control on deaminiting activity of yeast GDH and no control on reductive activity.

These results can be considered as a primary approach to elucidate the effect of oleanolic acid as yeast anaerobic growth factor: metabolic opportunities are not so numerous in anaerobiosis as they are in aerobiosis; the limited control of the deaminating activity of yeast GDH by oleanolic acid can maintain glutamic acid in a fairly steady state within the pool of intracellular aminoacid; thus in anaerobic condition oleanolic acid could participate in the activation process of protein synthesis.

- 1) P. BRECHOT et al., C. R. Ac. Sciences de Paris, 1971, 272, série D. 890-893.
- G. COHEN, Le Métabolisme cellulaire et sa régulation. Herman Edit., Paris 1967, p. 133.
- 3) J.B.C., 1965, 240, p. 3795.
- 4) GDH from Bovine liver, Sigma Chemical Company, type II, Crystalline, Sodium Phosphate Glycerol solution, free of Ammonium ions.
- 5) We are grateful to Mrs. FOURCADE and Miss VENARD for a purified sample of yeast GDH, suspension in phosphate 0,1 M buffer 7.6 (Laboratoire de Biologie Physico-Chimique, Faculté des Sciences d'Orsay Prof. TONNELAT).

XVI. Department of Microbiology, Shizuoka College of Pharmacy, 2-2-1 Oshika, Shizuoka, Japan. Communicated by Tamotsu Morita.

The following is a summary of a recent paper.

Action of 4-Nitroquinoline 1-Oxide on Mitochondrial DNA in Yeast.

Tamotsu Morita and Ichiji Mifuchi.

GANN 65(1), 27-32, 1974.

SUMMARY

To study the action of a carcinogen, 4-nitroquinoline 1-oxide (4NQO), on yeast mitochondrial DNA, synchronous cell cultures of yeast were used. The induction rate of cytoplasmic responation-deficient (RD) mutants of yeast by 4NQO was closely correlated to the yeast cell cycles, especially to mitochondriogenesis, such as the number of mitochondria in the cells, activities of some respiratory enzymes in mitochondria, and the replication of mitochondrial DNA.

The hydroxyapatite column chromatography of DNA preparations from intact cells or mitochondria fraction of RD mutant strain N-1, which was induced by 24-hr treatment of normal yeast with 4NQO, showed that the peak of mitochondrial DNA in this mutant disappeared. The present work does not provide a definite clue to what kind of damage resulted from 4NQO treatment in mitochondrial DNA in yeast. However, this result may suggest several possibilities, such as loss of mitochondrial DNA and considerable change in molecular weight or GC content of mitochondrial DNA in yeast. In this respect, recent work in our laboratory by cesium chloride density gradient centrifugation has shown that mitochondrial DNA in RD mutant strain N-1 disappeared. Detailed studies on inhibitory mechanisms of 4NQO on the synthesis of mitochondrial DNA in yeast are now in progress.

XVII. Arbeitsgruppe Mikrobengenetik, Fachbereich Biologie, J.W. Goethe-Universität, 6 Frankfurt/Main, Siesmayerstr. 70, Germany. Communicated by M. Brendel.

> Since the last issue of the Yeast News Letter the following two papers have been accepted for publication:

Economizing DNA-specific Labelling by Deoxythymidine-5'-monophosphate in <u>Saccharomyces</u> cerevisiae. Wolfgang W. Fäth, Martin Brendel, Wolfgang Laskowski and Elke Lehmann-Brauns. Molec. Gen. Genetics, in the press.

Summary. Although strain 211-laM is able to take up 5'-dTMP it cannot overcome growth-inhibition caused by an aminopterin (APT)-sulfanilamide (SAA)- mediated block of thymidylate biosynthetis in the presence of exogeneous 5'-dTMP. Eleven mutants (T-strains) of strain 211-laM were isolated being able to grow under these conditions.

Genetical analysis of the T-strains revealed in each case the mutation of a single recessive gene, TYP, to be responsible for this character, and that the mutated genes belong to three complementation groups.

One such <u>typ</u>-mutant, strain 211-laMT2, studied in detail was found to utilize exogeneous 5'-dTMP as poorly as strain 211-laM. However, when thymidylate-biosynthesis is inhibited by APT plus SAA the utilization of 5'-dTMP is increased by a factor 2.5 as compared to conditions where the antimetabolites are absent.

Next we isolated a typ-mutant- derivative (typ tlr- mutant) utilizing exogeneous 5'-dTMP better by a factor 2.5 than strain 211-laM2. This typ tlr- mutant was made auxotrophic for 5'-dTMP. The efficiency of utilization of exogeneous 5'-dTMP found for these typ tlr tmp-mutants was the same as that of their typ tlr- parental strain in the presence of APT plus SAA.

"Thymineless Death" in a Strain of Saccharomyces cerevisiae
Auxotrophic for Deoxythymidine-5' -monophosphate. Martin
Brendel and Ursula Langjahr, Molec. Gen. Genetics, in the press.

Summary. Cells of Saccharomyces cerevisiae strain 211-latmpl-1 which are auxotrophic for 5'-dTMP exhibit "thymineless death" (TLD) when deprived of the nucleotide. After an initial lag of about one generation time cells lose viability in exponential fashion halving their titer every 90 min. Thymine and thymidine (100 µg/ml) cannot prevent TLD in the absence of 5'-dTMP. Although the cell titer is constant during 24 hrs of 5'-dTMP deprivation, the cell mass synthesized increases by a factor of six during this period. Budding is stopped and cells attain a swollen shape. Synthesis of RNA and protein does occur in cells deprived of 5'-dTMP, the rates of synthesis being significantly lower than those in the controls.

XVIII. Genetics of Induction and Catabolite Repression of Maltase Synthesis
in Saccharomyces cerevisiae. Mykologie/Genetik, Technische Hochschule,
61 Darmstadt, Schnittspahnstr., German Federal Republic. Communicated
by F. K. Zimmermann.

Synthesis of maltase in yeast requires induction by maltose, and absence of glucose, catabolite repression. Induction also requires the presence of any one of the six known MAL-(maltose)-genes. In the absence of these genes, there are only trace amounts of maltase to be found. Two of the known MAL-genes, MAL4 and MAL6, have been shown to be regulatory genes. As reported previously, I have obtained two classes of mutant alleles of the gene locus MAL2, constitutive but glucose sensitive ones, and constitutive and glucose resistant alleles.

Further analysis has established that the regulatory effects of these alleles extend to the synthesis of alpha-methylglucosidase even in the absence of the known MGL-(alpha-methylglucoside)-genes. Another enzyme involved in carbohydrate catabolism is invertase which is repressed on glucose media. A possible effect of one of the glucose resistant constitutive alleles, MAL2-47°, on glucose repression of invertase was studied using the two SUC-(sucrose)-genes SUC2 and SUC3. Invertase specific activities were compared in strains with SUC2 and SUC3 to strains carrying these genes in combination with MAL2-47° after growth on a YEP-medium with 0.3% glucose and 1% glycerol (de-repressing medium) and on YEP-8% glucose (repressing medium). Maltase synthesis was repressed about 90% under these conditions, but it was still about 50 times higher than in wild type grown under less repressing conditions. Invertase synthesis was repressed in the SUC2 strain with MAL2-47° 90%, and 93% without MAL2-47°. Repression in the SUC3 strain was 99.7 versus 99.4%. MAL2-47° does not influence regulation of invertase synthesis, its range of action is restricted, it is not a general gene regulating catabolite repression.

MAL2 seems to be a positively regulatory gene because negative mutants, mal2, are recessive to MAL2, and constitutive as well as constitutive-catabolite repression resistant alleles are dominant at the level of maltase specific activities in respect to constitutivity. In contrast to this, resistance to catabolite repression is more variable in various allelic combinations. There are allele specific interactions notably with a negative mutant allele, mal2-4. A MAL2-47^c/mal2-4 diploid grown on 2% glucose YEP has only 10% of the maltase spec. activity found in a MAL2-47^c/MAL2-47^c diploid grown on 2% glucose YEP. This level, however, is still 30 times that of repressed wild type.

The most intriguing aspect of mutants like MAL2-47^c is that they suggest that catabolite repression is largely (not exclusively though) exerted in the positive control system rather than at the promoter site of the (as yet unidentified) structural gene(s) for maltase. This interaction could occur at the level of the regulatory gene product as suggested by the allele specific dominance levels observed in various heteroallelic diploids.

Genetic tinker toys: I have constructed a strain, D7, a diploid of the genotype: ade2-40/ade2-119 (this allows to study mitotic crossing over, red/pink twin spots), trp5-12/trp5-27 (to study mitotic gene conversion, convertants grown on tryptophan-less media) and ilv1-92/ilv1-92 (to study reverse mutation, reverse mutants grow on media without isoleucine). The advantage of D7 is that three different genetic events resulting from mutagenic treatment can be assayed on the same cell population. Coincidence between gene conversion at the trp5 locus on chromosome VII, mitotic crossing over on chromosome XV, and reverse mutation at the ilv1 locus on chromosome V and other suppressor mutations at as yet unknown locations can be followed. Mutant allele ilv1-92 can be reverted with a wide spectrum of mutagens.

XIX. Brooklyn College, Department of Biology, The City University of New York, Brooklyn, New York 11210. Communicated by Norman Eaton.

Michael Waxman has completed his doctoral dissertation entitled "On the Nature of the Control of the Transmission and the Expression of Mitochondrial Antibiotic Resistance Factors in Saccharomyces cerevisiae," an abstract of which follows:

The results of this study clearly demonstrated that nuclear factors can control the distribution of mitochondria from zygotes to their daughter cells. All methods of analysis used (random diploids, zygote clone and zygote cell lineage analysis) attested to this fact. The inhibition of nuclear RNA transcription and/or cytoplasmic protein synthesis in young zygotes for 90 minutes altered the preferential transmission of mitochondrial types to zygote daughter cells which was observed in untreated zygotes. Studies in which mitochondrial protein synthesis was inhibited with antibacterial antibiotics indicated that mitochondrial protein synthesis played little or no part in the events leading to recombination of mitochondrial DNA and subsequent transmission of mitochondrial resistance factors to the descendants of the zygotes. Genetic analysis of the cellular location of the control function showed that it resided in the nuclear genome, since the control function showed a Mendelian segregation pattern (2:2) at meiosis. The analysis of suppressive petite mutants in crosses to strains carrying the nuclear control genes has shown that these control genes can alter the suppressiveness of the suppressive petite mutants to levels approaching that of the neutral petite phenotype. The control function responsible for the asymmetrical distribution of mitochondrial resistance factors and suppressiveness are not, however, manifestations of a common mechanism but are two independent mechanisms which can be additive in their effects. The analysis of the omega factor and its effects in crosses to strains possessing the nuclear control determinants indicated that the omega factor did not modify or alter the asymmetrical inheritance pattern of strains carrying nuclear control genes.

James Paterniti, D. Wilkie and N. R. Eaton: Progress report on some studies on the selective inhibition of the growth of Saccharomyces by the proline analogue Thiazolidine-4- carboxylic acid and its probable modes of action:

The study of thiazolidine-4-carboxylic acid (TZ) was originally begun in our laboratory as part of a program to screen amino acid analogues in an attempt to find analogues which would be incorporated specifically into mitochondrially synthesized protein. Although the status of TZ as an analogue of this type is in doubt, much work on its properties and modes of action in yeast has been done. Cells of the haploid strain, 1493-10C, grown in 0.5% yeast extract (YE) + 2% glucose + 10 mg/ml TZ show a characteristic growth lag lasting from 8-10 hours, after which log phase growth proceeds at essentially untreated rates. The same overall growth pattern is seen when 0.5% YE + 3% glycerol or 0.5% YE + 0.5% acetate media are used in the presence of 10 mg/ml TZ. By contrast, growth is completely inhibited by

5 mg/ml TZ when 0.5% YE + 3% ethanol or 0.5% YE + 3% lactate are the growth media. Proline added to the culture medium in suitable amounts reduces the lag on glucose + TZ medium but does not reverse the inhibition of growth in ethanol + TZ or lactate + TZ cultures. In addition, acriflavin-induced petites of 1493-10C also show a typical lag on glucose + TZ medium. This observation, together with the selective proline reversal of TZ inhibition, suggests different modes of action for TZ in glucose and ethanol media. Studies on alcohol dehydrogenase from 1493-10C indicate that TZ is a non-competitive inhibitor of this enzyme. This observation probably explains inhibition of growth on ethanol + TZ medium. Studies using cells grown on YE + glucose + $^{14}\mathrm{C}\text{-TZ}$ indicates that the label is incorporated into protein. Furthermore, uptake studies show that glucose-grown cells exclude TZ as the lag phase of growth progresses. These observations partially explain lag and recovery of cells grown on glucose + TZ medium. Work in progress at this time centers around genetic and biochemical characterization of two classes of mutants: One class is resistants to TZ when grown on ethanol: The other class of mutants is sensitive to TZ when grown on glucose.

Brief report on alpha-methylglucosidase formation: Strains carrying any of the dominant MAL genes (MALXmg1) can ferment maltose (Ma) but not alpha-methylglucoside (Mg). Induction of such strains with either Ma or Mg results in the formation of both maltase (M'ase) and alpha-methylglucosidase (MG'ase I) in a ratio of about 5-10 to 1, although the levels of the enzymes produced by Mg induction are about 1/10 those with Ma and do not permit Mg fermentation. In strains also carrying MGL1 or MGL3, complementation resulting in Mg fermentation occurs only with MAL3(MGL2) or MAL4. In cells with the genotype MAL3/MGL1, Mg (but not Ma) induces an additional MG'ase (MG'ase II) which can be separated from MG'ase I by iso-electric focusing. In cells of the other complementing genotype (MAL4/MGL1) Mg induces only M'ase and MG'ase I. Induction of MG'ase II, therefore, requires MAL3 (or MGL2) in addition to MGL1.

The following papers have been published or accepted for publication:

Waxman, M. F., Eaton, N. and Wilkie, D., Effect of antibiotics on the transmission of mitochondrial factors in <u>Saccharomyces</u> cerevisiae, Molec. gen. Genet., 127:277 (1973).

Neddleman, R. and Eaton, N. R., Selection of yeast mutants constitutive for maltase synthesis, Molec. gen. Genet., in press.

XX. National Institutes of Health, National Institute of Arthritis,

Metabolism, and Digestive Diseases, Building 4, Room 116, Bethesda,

Maryland 20014. Communicated by Reed Wickner.

Below follow abstracts of two recent papers.

"Killer Character" of <u>Saccharomyces</u> <u>cerevisiae</u>: "Curing" by Growth at Elevated Temperature.

J. Bacteriol. 117, 1356-1357 (1974)

Normal "killer" strains of <u>Saccharomyces cerevisiae</u>, when grown at 37-40 C, produce almost exclusively non-killer cells due to loss or mutation of at least part of the non-chromosomal killer genome.

Chromosomal and Nonchromosomal Mutations Affecting the "Killer Character" of Saccharomyces cerevisiae.

Genetics, March 1974.

The "killer character" of <u>Saccharomyces</u> <u>cerevisiae</u> is a nonchromosomal genetic element which imparts to cells carrying it (a) the ability to kill cells which lack it, by secreting the soluble macromolecular killer substance and (b) the ability to resist the killing of the killer substance.

Mutants have been isolated from <u>S. cerevisiae</u> carrying the "killer character". Mutants were found in two nuclear genes (mak 1 and mak 2) involved in maintenance of the genetic element of the killer character. Mutants in three other nuclear genes could maintain the cytoplasmic genetic element normally; two of these were able to express resistance to killing, but not the ability to kill (kex 1 and kex 2), while the third could express the killing function, but was sensitive to killing and is thus a "suicide" strain (rex 1). These five nuclear genes were all distinct.

Several mutants were detected which showed non-Mendelian segregation indicating mutation of the "cytoplasmic" genetic element. Some such mutants had normal resistance to killing but were either unable to kill at all or showed marked reduction in the ability to kill. Others had lost both killing ability and resistance. None were suppressive.

Two nonkiller mutants, both of which showed cytoplasmic inheritance, were mated. A large, but variable, proportion of diploid colonies, plated after several generations of growth, showed normal killing which was then stably inherited on further subcloning.

XXI. Department of Genetics, Haryana Agricultural University, Hissar-125004, Haryana, India. Communicated by R. K. Vashishat.

The following is a summary of work done in this department on UV-sensitive mutants of yeast.

Isolation and characterization of ultraviolet-sensitive mutants of Saccharomyces cerevisiae.

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R. K. Vashishat and S. N. Kakar

Twelve UV-sensitive mutants (UVS₁ to UVS₁₂) have been isolated. Four mutants were petites (two cytoplasmic, one nuclear and one did sporulate when crossed to wild-type). Genetic analysis of ten mutants has revealed that mutations affecting UV-sensitivity were single gene mutations, recessive and constituted seven complementation groups. Only one group was represented by four mutants.

The wild-type and five mutants showed decrease in survival in the presence of caffeine (0.1%) in the post-irradiation medium while two strains were not affected. These results show that in yeast some steps, but not all, of the dark-repair system are blocked by caffeine.

All the UV-sensitive and the wild-type strains showed increase in UV-survival after photoreactivation.

When tested for cross-sensitivity to X-ray, nitrous acid (HNO₂), ethyl methanesulfonate and N-methyl-N'-nitro-N-nitro-soguanidine (NG), strains UVS₁, UVS₈ and UVS₁₁ were sensitive to all the mutagens. Strains UVS₃, UVS₅ and UVS₉ were resistant to X-ray and NG but sensitive to UV, HNO₂ and EMS, except UVS₃ which was HNO₂ resistant. This shows that repair of radiation damage and damage caused by chemical mutagens has certain steps in common. UV-sensitive mutants of yeast have been classified into three groups comparable to HCR, REC and EXR mutants of E. coli.

UV-induced reversions to prototrophy at $ade\ 2-1$ and $his\ 5-2$ loci in repair-deficient strains (UVS $_3$, UVS $_4$, UVS $_5$ and UVS $_8$) and in a wild type strain have shown that in UVS $_3$ and UVS $_5$, the frequency of UV-induced reversions increased as compared to wild-type strain, while it was decreased in UVS $_4$. However, strain UVS $_8$ did not show any difference as compared to wild-type. Photoreactivation in all the strains decreased the frequency of UV-induced reversions indicating that in these strains the premutational damage is due to the formation of pyrimidine dimers.

Studies with UV-sensitive strains have lead us to conclude that (a) in yeast UV-induced mutations originate as errors in the recombinational repair of single strand gaps (b) the repair mechanisms and the mechanisms of UV-induced mutation in yeast are similar to those in E. coli.

XXII. Allied Breweries (Production) Limited, The Brewery, Station Street, Burton-on-Trent DE14 1BZ. Communicated by D. A. Lovett.

The following are abstracts of five recent papers written by members of the Process Research Department.

The Rapid Detection of Infection

J. Harrison, T.J.B. Webb, P. A. Martin

Paper presented to the Annual Convention American Society of Brewing Chemists, April 1974.

It has been found possible to detect small number of microorganisms, including <u>Saccharomyces carlsbergensis</u>, <u>S. cerevisiae</u>, <u>S. ellipsoideus</u> and <u>S. diastaticus</u>, in beer by measuring changes in the conductance or pH of the medium during incubation. These methods allow the presence of infecting micro-organisms to be detected more rapidly than do the trasitional methods.

Relationship between Melibiose Fermentation and Antigenic Behaviour of Saccharomyces Species

M. J. Chilver, T.J.B. Webb, P. A. Martin

Paper to be presented to the 4th International Symposium on Yeasts, Vienna, July 1974.

From mating strains of Saccharomyces cerevisiae and S. carlsbergensis 195 stable hybrids, each the result of S. cerevisiae x S. carlsbergensis cross, have been produced. The antigenic reaction pattern for each of these hybrids and of the mating strains from which they were derived, their abilities to ferment melibiose and the relationship between antigenic behaviour and melibiase activity have been examined. Of the 195 hybrids examined, 161 (83%) gave the S. carlsbergensis Type II antigenic response and were able to ferment melibiose. Only 3 (1.5%) gave the S. cerevisiae antigenic response combined with inability to ferment melibiose. None gave the S. cerevisiae antigenic response combined with the ability to ferment melibiose and 100% of the hybrids that fermented melibiose gave the S. carlsbergensis Type II antigenic pattern. However, 14 (7%) gave the S. carlsbergensis Type II antigenic pattern and yet were unable to ferment melibiose. Sixteen hybrids (8%) showed completely anomalous results antigenically. It is concluded that although there is a close relationship between melibiase activity and S. carlsbergensis Type II antigenic behaviour, the same site is not responsible for each factor.

The Production of Hydrogen Sulphide by Yeast and by Zymomonas anaerobia.

R. J. Anderson, G. A. Howard

J. Inst. Brew., 1974, 80, (May/June)

Radiochemical experiments indicated that most of the free hydrogen sulphide excreted by a brewing yeast in wort and by Zymomonas anaerobia in beer was derived from sulphate. Sulphate was also assimilated by the organisms but most of their cellular sulphur was derived from other sources. In a synthetic beer medium, excessive amounts of hydrogen sulphide were liberated by \underline{Z} . anaerobia when pantothenate was deficient. Sulphate and zinc ions stimulated sulphide production by the bacterium.

The Origin and Occurrence of Volatile Sulphur Compounds in British Ales and Lagers.

R. J. Anderson, G. A. Howard

J. Inst. Brew., 1974, in press

A flame-photometric sulphur detector was used to identify, measure and determine the sources of the sulphur volatiles produced during the commercial processing of British ale and lager. Malt and hops contained a number of sulphur volatiles, but most of this material extracted into commercial worts was driven off duirng boiling. Brewing yeasts produced only traces of organo-sulphur volatiles both in laboratory fermentations of wort and during the processing of commercial ales and lagers. Brewery bacteria, particularly wort spoilage organisms, could generate sulphur volatiles.

The Rapid Detection of Brewery Micro-Organisms

J. Harrison, T.J.B. Webb, P. A. Martin

J. Inst. Brew., 1974, in press

Recent developments in the rapid detection of low concentrations of micro-organisms are discussed and their relevance to improved brewery quality control methods is assessed. A method based on the change in pH of a general purpose growth medium appears to offer the advantages of both time and sensitivity over methods currently in use. Saccharomyces cerevisiae is among the organisms studied by the pH change method.

XXIII. Miller Brewing Company, Milwaukee, Wisconsin 53201. Communicated by J. F. Rice.

Below follows the abstract of a paper I presented at the convention of the American Society of Brewing Chemists, in San Francisco entitled "The Quantitative Influence of Agitation on Yeast Growth during Fermentation".

The literature indicates in qualitative terms that agitation during fermentation increases the amount of yeast grown. We have quantified this relationship and have established that the magnitude of both maximum growth and maximum growth rate are directly related to the degree of turbine-type agitation, but is inversely related to the concentration of carbon dioxide in fermenting wort. We therefore hypothesize that agitation influences maximum growth and maximum growth rate through feedback inhibition by carbon dioxide.

This hypothesis not only explains the observed influence of turbine-type agitation on maximum growth and maximum growth rate, but also explains two other observations---viz., (1) high maximum growth and maximum growth rate even with relatively gentle gyrorotatory agitation and (2) reduced maximum growth at reduced temperatures. Although the use of unmodified

Erlenmeyer flasks on a gyrorotatory shaker provides a high degree of laminar flow, it also provides highly efficient removal of dissolved carbon dioxide. In the second case the increased solubility of carbon dioxide at reduced temperatures can reasonably explain the reduced growth.

XXIV. Horticultural Products Laboratory, Horticultural Research Institute of Ontario, Vineland Station, Ontario LOR 2EO, Canada. Communicated by R. V. Chudyk.

The following has been published:

Chudyk, R. V. (1973) Media for isolating Ontario winery microflora. J. Inst. Brew. 79:509-512.

Abstract

Twenty-one microbiological media were evaluated statistically as isolation media for airborne microflora in an Ontario winery bottling room. Settleplate analysis indicated that different media varied in their ability to isolate numbers and types of viable microorganisms. YM Agar (pH 6.2) yielded the highest total as well as the highest yeast counts. YM Agar (pH 6.2) and Sabouraud Medium (pH 6.2) yielded the highest mould counts. Micro Assay Culture Agar (pH 6.7), Brewer Anaerobic Agar (pH 7.2) and a modified YM Agar (pH 7.0) yielded the highest bacterial counts. There was no exclusive relationship between the viable count and pH on Malt Extract Agar, Sabouraud Medium, Wort Agar, W. L. Nutrient Medium and YM Agar.

XXV. University of Rochester, School of Medicine, Rochester, N.Y. 14642. Communicated by Fred Sherman.

Excerpt from: F. Sherman and C. W. Lawrence, <u>Saccharomyces</u> In "Handbook of Genetics" vol. 1, edited by R. C. King, Plenum Pub. Corp. in press.

GENETIC NOMENCLATURE

A recommendation for the nomenclature used in yeast genetics was published as the November 1969 supplement to the Microbial Genetics Bulletin, and except for the persistence of a few of the older notations, these proposals are usually followed. For the sake of clarity and convenience, a few minor changes appear appropriate and these are included in the following rules of nomenclature which have been expanded and updated:

- 1. The two mating type alleles are designated, respectively by a and α . (In the old system, the itacilized <u>a</u>, in contrast to the bold face <u>a</u>, was sometimes difficult to differentiate from α .)
- 2. Gene symbols are usually designated by three italicized letters which should be consistent with the proposal of Demerec et al., (1966) whenever applicable, e.g., arg. The genetic locus is identified by a number immediately following the gene symbol, e.g., arg2. Alleles are designated by a number separated from the locus by a hyphen, e.g., arg2-6. While locus numbers must agree with the original assignments, allele numbers may be particular to each laboratory.
- 3. Complementation groups of a gene or a gene cluster can be designated by capital letters following the locus number, e.g., his4A, his4B, etc.
- 4. Dominant and recessive genes are denoted by upper and lower case letters, respectively, e.g., SUP6 and arg2.
- 5. When there is no confusion, wild-type genes are designated simply as +; the + may follow the locus number to designate a specific wild-type gene, e.g., sup6+ and ARG2+.
- 6. While superscripts should be avoided, it is sometimes expedient to distinguish genes conferring resistance and sensitivity by the superscript R and S, respectively. For example the genes controlling resistance to canavanine sulfate (can1) and copper sulfate (CUP1) and their sensitive alleles could be denoted, respectively, as can^R1, CUP^R1, CAN^S1 and cup^S1.
- 7. Mitochondrial and other non-Mendelian genotypes can be distinguished from the chromosomal genotype by enclosure in square brackets. Whenever applicable, it is advisable to employ the above rules for designating non-Mendelian genes, and to avoid the use of Greek letters; however it is less confusing to either retain the original symbols ρ^+ , ρ^- , ψ^+ and ψ^- or to use their transliteration, respectively, [rho+], [rho-], [psi+] and

[psi-]. The mitochondrial genes conferring resistance and sensitivity can be denoted by superscripts as indicated in (6). Since the identification of distinct genetic loci and the dominance relationships of alleles are less clear, genes can be denoted by capital letters and mutant numbers, e.g., [ERYS CHLR 321], [ERYR 221 CHLS], etc.

XXVI. International and other meetings.

Fourth International Symposium on Yeast and other Protoplasts. Department of Botany, University of Nottingham, U.K., September 8-12, 1975.

Organizing Committee: Professor E. C. Cocking, Nottingham - Chairman; Dr. J. F. Peberdy, Nottingham - General Secretary; Professor P. Matile, Zurich; Dr. H. J. Rogers, London; Professor A. H. Rose, Bath.

Secretariat: Department of Botany, University of Nottingham, University Park, Nottingham NG7 2RD, U.K.

The Meeting will be held in the Department of Botany, University of Nottingham. The official language of the Symposium will be English.

Scientific Programme:

- A. Plenary Lecture.
- B. The following topics have been selected for the main sessions of the Symposium.

 Where possible, papers will be presented to show comparative aspects of protoplast work.

Isolation and properties of protoplasts
Uptake and viral interactions: transformation and transduction
Fusion and somatic hybridization
Cell walls and membranes - biosynthetic capabilities
and growth.

C. Free communications on the above topics.

TENTATIVE SCHEDULE

Monday, September 8th

Opening session, with evening Plenary Lecture. Welcome Reception.

Tuesday, September 9th

a.m. Symposium sessions

p.m. Free communications

Wednesday, September 10th

a.m. Symposium sessions

p.m. Excursion

Thursday, September 11th

a.m. Symposium sessions

p.m. Free communications

Friday, September 12th

a.m. Symposium sessions

p.m. Free communications

Conference Dinner

Second Announcement

This First Announcement contains a preliminary form, which should be filled in and returned before August 1, 1974, by all those wishing to receive the Second Announcement. The latter will contain the preliminary programme, registration and accommodation forms, abstract reproduction forms, etc.

COLLOQUIUM ON GENETICS AND BIOGENESIS OF CHLOROPLASTS AND MITOCHONDRIA

A colloquium on "Genetics and Biogenesis of Chloroplasts and Mitochondria" will be held at The Ohio State University, Columbus, Ohio on September 5-7, 1974, sponsored by the College of Biological Sciences. Presentations will cover mitochondrial and chloroplast genetics and biogenesis in microbial, plant, and tissue culture systems; mitochondrial mutagenesis; the use of organelle genes in analysis of oxidative phosphorylation and in plant breeding; and the role of organelle genes in evolution. Speakers are G. Attardi. C. W. Birky, Jr., D. E. Griffiths, J. K. Hoober, J. Laughnan, H. R. Mahler, P. S. Perlman, R. Sager, R. A.E. Tilney-Bassett, and S. G. Wildman.

For information about registration write to:

COLLOQUIUM College of Biological Sciences The Ohio State University Columbus, Ohio 43210

XXVII. Brief News Items:

1. Dr. Yasuo Umeda of the Kirin Brewery Ltd. writes: I have pleasure in informing you that I retired from the post of Senior Managing Director and have been appointed Standing Advisor to the Company.

My responsibility at this new post includes the management of the Research Laboratory of the Kirin Brewing Co. (Takasaki, Gumma Pref. Japan) following the transference of Dr. Yoshiro Kuroiwa to the Takasaki Brewing Plant.

- 2. Mr. Tatsuhiko Kaneko, Technical Information Section, The Research Laboratories of the Kirin Brewing Co. Ltd, Takasaki, Gumma Pref., Japan, writes:
- 1. In April 1974, Dr. Yasushi Yamamoto was transferred to the Sendai Brewing Plant of our company.
 - 2. Recent publication:
 - T. Kaneko, K. Kitamura and Y. Yamamoto Susceptibilities of Yeasts to Yeast Cell Wall Lytic Enzyme* of <u>Arthrobacter luteus</u>. Agr. Biol. Chem., 37, 2295 (1973).

*This enzyme preparation is being sold by our Laboratories.

3. Dr. F. Schlenk, Division of Biological and Medical Research, Argonne National Laboratory, 9700 South Cass Ave., Argonne, Illinois 60439, USA writes:

I am trying to place my associate, Dr. K. D. Nakamura, whose stipend here as a Postdoctoral Research Associate is coming to an end; there is a two years' mandatory limit of tenure. I cannot make other arrangements here for Dr. Nakamura, because my activities will come to an end with my retirement, late in 1974.

Dr. Nakamura's work at Argonne has been concerned with the isolation and properties of yeast cell vacuoles, radiation damage, budding defects, and related biochemical aspects; several papers are in press. Incidental to this, Dr. Nakamura has developed an ususual proficiency in ultraviolet photography.

Dr. Nakamura's educational background is as follows and he may be contacted at the above address:

Undergraduate: B. S. 1967. University of California
Berkeley, California
Major: Chemistry

Graduate: M.S. 1969. Oregon State University
Corvallis, Oregon
Major: Microbial physiology and genetics

Ph.D. 1972. Oregon State University
Corvallis, Oregon
Major: Microbial physiology and Biochemistry
Thesis title: Aspects of the regulation of
methyl group formation in
methionine biosynthesis by
Saccharomyces cerevisiae.

Professional Experience.

Postdoctoral Appointment: 1972 to present. Division of Biological and Medical Research Argonne National Laboratory
Argonne, Ill. 60439

Dr. Fritz Schlenk, sponsor.

- 4. Dr. W. Ch. Slooff, formerly with the CBS culture collection in Delft, the Netherlands and now retired, informed the Editor that an article by P. J. Nieuwdorp, P. Bos and W. Ch. Slooff, dealing with the systematics, capsular composition and ascospore properties of <u>Lipomyces</u>, has been accepted for publication in Antonie van Leeuwenhoek. This article was originally referred to in The Yeasts (J. Lodder ed.) as Nieuwdorp <u>et al.</u> 1970.
- 5. Late last fall Professor and Mrs. Carl C. Lindegren celebrated their 50th Wedding Anniversary. Your many friends in the Yeast World extend their warmest congratulations!

The Lindegren team has submitted the following publications for inclusion in the Yeast News Letter.

- Carl C. Lindegren The Mitochondria as Vehicles of Entry and of Transport. Jr. Biolog. Psych. 13:355, 1971.
- Carl C. Lindegren Cellular Organization in Relation to Evolution. Jr. Biolog. Psych. 14:38-43, 1972.
- Carl C. Lindegren and Gertrude Lindegren Mitochondrial Modification and Respiratory Deficiency in the Yeast Cell Caused by Cadmium Poisoning. Mutation Res. 21:315-322, 1973.
- Carl C. Lindegren and Gertrude Lindegren 1973. Oxidative detoxication of thallium in the yeast mitochondria. Antonie v. Leewen. 39:351-353. Southern Illinois University, Carbondale, Illinois.
- 6. Dr. N. J. W. Kreger-van Rij, Oostersingel 59, Groningen (Holland) writes:

Below follow two recent publications

- N. J. W. Kreger-van Rij and M. Veenhuis, Electron microscopy of septa in ascomycetous yeasts. Antonie van Leeuwenhoek, 39: 481-490, 1973.
- N. J. W. Kreger-van Rij and M. Veenhuis, Spores and septa in the genus Dipodascus. Can. J. Bot. (in press).

7. I would like to inform the readers that the following paper will appear in the January issue, Vol. 27, No. 1 of Microbiologia Española (Madrid).

Ramirez, C. 1974. A Compilation of description of New <u>Candida</u>
Species with keys to all species of the genus described up to date.

This paper will contain standard descriptions of the new species of <u>Candida</u> that have been discovered in the world up to date after the coming up of the second revised edition of Lodder's The Yeasts (1970), and keys to all species of <u>Candida</u>.

Prof. Dr. C. Ramirez
Consejo Superior de Investigaciones
Cientificas
Instituto Jaime Ferran de
Microbiologia
Joaquin Costa, 32
Madrid, 6 (Spain)