Y E A S T

A News Letter for Persons Interested in Yeast

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I. Announcements by the Editor

The Editor wishes to extend to all readers of the Yeast News Letter a prosperous and scientifically rewarding year.

Because of considerable increases in postal rates and other costs the subscription price of the Yeast News Letter has been increased from $1.50 to $2.00 per year. The increase is reflected in the bill which accompanies this issue of the News Letter. Individuals who have paid the 1976 subscription last year will be excused from the price increase. Furthermore, all subscribers who have not paid for two years will be dropped from the mailing list.

Erratum: The volume and year of the Spring issue of the Yeast News Letter of 1975 should be changed from Vol. XXIII to XXIV and the year from 1974 to 1975.

H. J. Phaff

II. Centraalbureau Voor Schimmelcultures (Netherlands), Yeast Division, Delft, Julianalaan 67A. Communicated by David Yarrow.

Below follows a list of new species received by the CBS since my last contriubtion to the newsletter.

Candida brassicae CBS 6799 = RIFY E-17, Type; Y. Amano, S. Goto & M. Kagami, J. Ferment. Technol. 53: 311-314 (1975).

Candida buinensis CBS 6796 = IFO 1642, Type
Pichia rabaulensis CBS 6797 = IFO 1643, Type
Schizoblastosporion kobayasii CBS 6798 = IFO 1644, Type
M. Soneda and S. Uchida, Bull. Nat. Sci. Mus. Tokyo 14: 138-459, 1971

Candida flavificans CBS 6735 = AJ 4997, Type
Candida inositophilia CBS 6736 = AJ 5000, Type
Torulopsis sorbophila CBS 6739 = AJ 4995, Type
T. Nakase, A. van Leeuwenhoek 41: 201-210 (1975)

Candida hydrocarbofumarica CBS 6734 = AJ 5204
K. Yamada, T. Furukawa & T. Nakahara, Agr. Biol. Chem. 34:670-675 and 1402-1406 (1970)

Candida savonica CBS 6563, Type
C. E. Sonck, A. van Leeuwenhoek 40: 543-545 (1974)

Cryptococcus cereanus CBS 6644 = UCD 73-313, Type; CBS 6645 = UCD 73-67. H. J. Phaff, M. W. Miller, M. Miranda, W. B. Heed & W. T. Starmer, Int. J. Syst. Bacteriol. 24:486-490 (1974).

Endomycopsis montevidensis CBS 6721 = IMUR 2329, Type
L. A. de Queiroz, Mycopath. Mycol. Appl. 51:307314 (1973)

Hanseniaspora occidentalis CBS 2592, Type; CBS 6782, CBS 6783
M.Th. Smith, A. van Leeuwenhoek 40: 441-444 (1974)

Hansenula lynferdii CBS 6695, Type
J. P. van der Walt & E. Johannsen, A. van Leeuwenhoek 41:13-16 (1975)

Lipomyces anomalus CBS 6740 - DSB 2573, Type
I. P. Babjeva & S. E. Gorin, A. van Leeuwenhoek 41: 185-191 (1975)

Pichia norvegensis CBS 6564, Type; CBS 6639
B. Leask & D. Yarrow, Sabouraudia, in press

Pichia philogaea CBS 6696, Type; CBS 6725
J. P. van der Walt, A. van Leeuwenhoek 41: 173-177 (1975)

Trichosporon arenicola CBS 6722 = IMUR 2330
S. M. de Lima and L. A. de Queiroz, Univ. Fed. Pernambuco Inst. Micol. Publ. 690: 1-8 (1972)

Trichosporon terrestre CBS 6697, Type
J. P. van der Walt & E. Johannsen, A. van Leeuwenhoek 41:361-365 (1975)

III. American Type Culture Collection, 12301 Parklawn Drive, Rockville, Maryland 20852. Communicated by D. S. King.

Below follows a list of strains accessioned to the ATCC since the list published in the Spring issue of the Yeast News Letter.

Complete information (isolation substrate, depositor, preferred media, etc.) will be available in the American Type Culture Collection Catalogue of Strains, 12th edition, which will be available for distribution in January of 1976.

Candida albicans
ATCC 28956, ATCC 32089

Candida diddensii
ATCC 28872

Candida guilliermondii
ATCC 28873

Candida krusei
ATCC 28870

Candida pseudotropicalis
ATCC 28908

Candida tropicalis
ATCC 32113

Candida utilis
ATCC 28955

Candida veronae
ATCC 28874

Cryptococcus laurentii var. magnus
ATCC 32065

Cryptococcus luteolus
ATCC 32044

Cryptococcus neoformans
ATCC 32045

Cryptococcus terreus
ATCC 32046

Cryptococcus uniguttulatus
ATCC 32047, ATCC 32048

Filobasidiella neoformans
ATCC 28957, ATCC 28958

Hanseniaspora occidentalis
ATCC 32053

Kloeckera brevis
ATCC 32076

Candida zeylanoides
ATCC 28871

Cryptococcus albidus
ATCC 32037, ATCC 32038, ATCC 32039,
ATCC 32040, ATCC 32041

Cryptococcus albidus var. albidus
ATCC 32036, ATCC 32059, ATCC 32060,
ATCC 32061

Cryptococcus albidus var. diffluens
ATCC 32062

Cryptococcus gastricus
ATCC 32042

Cryptococcus lactativorus
ATCC 32043

Cryptococcus laurentii var. flavescens
ATCC 32063

Cryptotoccus laurentii var. laurentii
ATCC 32064

Saccharomyces cerevisiae
ATCC 32049, ATCC 32050, ATCC 30251
ATCC 32052, ATCC 32119, ATCC 32120
ATCC 32145, ATCC 32146, ATCC 32147
ATCC 32148, ATCC 32167

Saccharomyces delbrueckii
ATCC 28916

Saccharomyces fermentati
ATCC 28917

Saccharomyces italicus
ATCC 28918

Saccharomyces rosei
ATCC 32035

Saccharomyces uvarum
ATCC 28919, ATCC 28920

Saccharomyces willianus
ATCC 28921

Sporobolomyces roseus
ATCC 28988

Kluyveromyces drosophilarum
ATCC 28909

Kluyveromyces fragilis
ATCC 28910

Kluyveromyces lactis
ATCC 28911, ATCC 32143, ATCC 32144

Kluyveromyces marxianus
ATCC 28912

Kluyveromyces veronae
ATCC 28913

Lipomyces lipofer
ATCC 32031

Rhodotorula citrinis
ATCC 28983

Saccharomyces bayanus
ATCC 28914, ATCC 28915

Sterigmatomyces nectairii
ATCC 32127

Sterigmatomyces penicillatus
ATCC 32128

Syringospora albicans
ATCC 32033

Syringospora stellatoidea
ATCC 32077

Torulopsis bacarum
ATCC 28954

Torulopsis multis-gemmis
ATCC 28953

Torulopsis pustula
ATCC 32034

Torulopsis sonorensis
ATCC 32108, ATCC 32109

Trichosporon oryzae
ATCC 32189

IV. Institute of Biochemistry and Physiology of Microorganisms of the USSR Academy of Sciences, Pushchino on Oka, Moscow region USSR. Communicated by W. I Golubev.

Recently Mrs. L. M. Vagabova and I have analyzed the nucleotide composition of DNA in some species of yeasts. The DNA extraction was done by the method of Schmidt and Thanhauser. Paper chromatography was used for the analysis of nucleotide composition (5-7 determinations for each DNA preparation). The GC content of Debaryomyces formicarius (CBS 6454) is 35.5 +0.4, Nadsonia elongata - 45.1 +0.8, N. commutata - 39.5 +1,1, Schizoblastosporion starkeyi-henricii - 42.2 ± 1.1, Torulopsis apis var. galacta nov. var. - 46.0 ± 0.5; Cryptococcus species - 52.4 ± 0.4, Cr. ater - 48.6 ± 1.1 Cr. himalayensis - 55.4 ± 0.8, Cr. hungaricus -56.5 ± 0.8; Cr. lactativorus (strain CCY-17-12-1) - 43.2 ± 1.0; Cr. macerans - 61.2 ± 0.7 and Cr. uniguttulatus - 51.5 ± 0.3 mol %.

The following articles and reports have recently been published:

Okunev, O. N., Golubev, W. I. 1974. Catabolism of i-inositol by cryptococci. VII. Interna. Sympos. on Carbohydr. Chem., Bratislava, Aug. 5-9, Abstr. 240.

Golubev, W. I., Okunev, O. N., Vdovina, N. V. 1974. The assimilation of i-inositol by yeasts as the diagnostic sign. Microbiologia, 43, N°6, 1046-1050.

Golubev, W. I., Okunev, O. N. 1975. Biochemical interpretation of diagnostic test to assimilate i-inositol in asporogenous yeasts. V All-Union Symp. Microbiol. Soc., Erevan, June 2-7, section "The Taxonomy of microorganisms," 59.

The following article is in preparation: Golubev, W. I., Bab'eva, I. P., Blagodatskaya, V. M. Reschetova, I. S. Taxonomic study of yeasts isolated from birch sap (Betula verrucosa Ehrh.).

V. Georgia State University, Department of Biology, Atlanta, Georgia 30303. Communicated by D. G. Ahearn.

The following papers have been published or are in press.

"Pichia Spartinae, A Dominant Yeast of the Spartina Salt Marsh." S. P. Meyers, D. G. Ahearn, S. K. Alexander and W. L. Cook. Deve. Ind. Microbiol. 16:262-267.

Abstract

Pichia spartinae, a salt-marsh yeast, occurs in concentrations as great as 9 X 10^7 cells/g in intraculm cell liquid and viable tissue of the estuarine angiosperm plant, Spartina alterniflora. Highest densities of the yeast occur on the outer surfaces of the plant during the summer and early fall. P. spartinae assimilates lipids of Spartina, plant hydrolysates, various amines, and both alpha-and beta-glucosides. An active β-glucosidase

system is indicated. Various plant residues and extracts support a significant cell crop. Little or negligible activity is noted against pentoses and soluble starch. Ascosporulation is observed most readily in fresh culm isolates and is frequently sparse in stock cultures. The unique association of this ascosporogenous yeast with the actively growing spartina plant suggests a relationship of notable environmental significance.

"Evaluation of the Uni-Yeast-Tek Kit for the Identification of Medically Important Yeasts." P. I. Bowman and D. G. Ahearn. J. Clin. Microbiol.

Abstract

The Uni-yeast-Tek system, a commercially prepared kit and scheme for the rapid identification of medically important yeasts (Corning Diagnostics), was evaluated in comparison with a conventional procedure in the identification of 623 yeasts. The system permitted the presumptive identification of 99.8% of 436 isolates representing 16 common species commonly isolated in the clinical laboratory. Correct biochemical and morphological analysis were obtained with 48 other species but their specific identification required additional data.

VI. Clinical Mycology Section, National Institute of Allergy and Infectious Diseases, NIH, Bethesda, Md. 20014. Communicated by K. J. Kwon-Chung.

Sexual Life Cycle of Cryptococcus neoformans

ABSTRACT

A basidiomycetous life cycle was found in Cryptococcus neoformans. When compatible pairs of isolates were inoculated on malt extract agar and incubated at temperatures between 15 to 37°C for 2 to 3 weeks, the sexual reproduction initiates with conjugation of two yeast cells which is followed by hyphal formation with clamp connections. Dikaryotic hyphae produce long basidia, singly or in a group, each having a subglobose to flask-shaped apex. Chains of sessile basidiospores are produced on the apex of each basidium by repetitious budding from four spots on the apex. Basidiospores are hyaline, ovate to globose and finely rough, measuring 1.8 to 2.5 µm. The perfect state of C. neoformans is described as Filobasidiella neoformans Kwon-Chung in the Filobasidiaceae of the Ustilaginales. Published in Mycologia, November - December issue of 1975.

VII. Ruhr-Universität Bochum, Institut für Physiologische Chemie, 4630 Bochum, Germany. Communicated by Wolfgang Duntz.

The following studies on the structure of α factor, the mating type specific sexual hormone of α cells of S. cerevisiae which specifically inhibits DNA synthesis and cell division in a cells, have been completed and will be published presently.

1. Heterogeneity of α factor.

Several compounds exhibiting α factor activity are found in culture filtrates of α cells. These compounds represent a group of related linear

peptides of 13 or 12 amino acids, designated as α_1, α_2, α_3, and α_4. The main compounds, α_1, and α_2, differ by an amino-terminal tryptophan residue which is present in α_1 and lacking in α_2. Otherwise the primary structures are identical. α_3 and α_4 are oxidation products of α_1 and α_2, respectively.

2. Primary structure of factor(s).

The following amino acid sequence has been found for α_1 and α_2:

H_2N-(Trp)-His-Trp-Leu-Gln-Leu-Lys-Pro-Gly-Gln-Pro-Met-Tyr-COOH. α_3 and α_4 are the methionine sulfoxides of α_1 and α_2.

3. Synthetic peptides with α factor activity.

Dieter Stotzler in our lab has synthesized several short peptides which represent part of the primary structure of α factor. A pentapeptide, H_2N-His-Trp-Leu-Gln-Leu-COOH, does not possess α factor activity by itself but the activity of native factor is increased by this peptide up to one hundred fold depending on peptide concentration. We assume that the pentapeptide either inhibits the degradation of native α factor or saturates non-specific binding sites for α factor on the surface of the a cells. The octapeptide of the structure

H_2N-His-Trp-Leu-Gln-Leu-Lys-Pro-Gly-COOH

possesses α factor activity, inhibiting cell division in a cells and causing the formation of characteristic "schmoos." Other research programs in our lab deal with the mechanism of action of α factor, the isolation and characterization of a factor and the study of a biologically inactive, α factor-like peptide produced by a sterile α strain carrying the mutation ste 1-2.

VIII. Laboratoire d'Enzymologie du C.N.R.S. 91190 Gif-sur-Yvette, France. Communicated by J. Schwencke.

The preservation of the vacuole content for electron microscopy in yeast protoplasts.

The yeast vacuole appears mostly as an electron-transparent area in the electron microscope. However, a dense, granular content can be observed in the vacuole, when protoplasts are fixed in iso-osmotic medium with 0.1 M glutaraldehyde and subsequently treated with 0.56 M glutaraldehyde. The vacuolar membrane can be visualized after a short post-fixation with 0.5% osmium oxide.

Protoplasts treated with 5% lead nitrate and 50% sodium sulfide, after the glutaraldehyde and osmium fixation, reveal the vacuole to contain an electron-dense precipitate evenly distributed. This precipitate may represent vacuolar polyphosphates. No other cellular structure contains this precipitate.

Isolated vacuoles can also be fixed by the same procedure, if the initial glutaraldehyde fixation is done in a vacuole-stabilizing medium. Their intravacuolar structure shows the same characteristics as those described for non-isolated vacuoles.

Our findings allow a clear differentiation between the yeast vacuole(s) and other electron-transparent structures.

(Abstract IVth International Symposium on Yeast and Other Protoplasts, Nottingham, England, September 1975).

Protoplasts of Schizosaccharomyces pombe. An advanced method for their preparation and the study of their guanine uptake. Pascal Housset and Maria Nagy.

Laboratoire de Génétique, Institut National Agronomique, 16, Rue Claude Bernard 75005 Paris, France, and Jaime Schwencke, Laboratoire d'Enzymologie du C.N.R.S., 91190 Gif-sur-Yvette, France.

Summary

A new method is described for the efficient conversion of S. pombe cells into protoplasts. The use of snail enzyme with added $\alpha(1\rightarrow3)$ glucanase and $\beta(1\rightarrow3)$ glucanase allowed the complete conversion of cells into protoplasts after 80 minutes of incubation.

The following parameters of guanine uptake determined in whole cells, were unchanged in protoplasts: Km value, requirement for an energy source, sensitivity to competitive inhibitors, pH optimum, as well as the typical variation of the initial velocity of uptake observed during the growth phase.

IX. Akademie der Wissenschaften der DDR, Zentralinstitut fur Mikrobiologie und Experimentelle Therapie, Jena. Communicated by H. Weber.

We present the summary of a recent paper by H. Weber and R. Lindner about "Virusartige Partikel in Hefeprotoplasten. I. Elektronenmikroskopischer Nachweis," soon to be published in Zeitschrift fur Allgemeine Mikrobiologie.

"Ultrathin sections and negatively stained isolates from protoplastsof the yeast S. cerevisiae with abnormal morphology and degeneration phenomena show the presence of viruslike particles within the cells. The isometric particles have diameters of about 50 nm, the electron dense centre is surrounded by a distinct envelope. Protoplasts containing up to 30,000 particles can be found. It could be demonstrated that there exists a correlation between the amourt of particles and the degree of degeneration of substructure. Protoplasts with high titers of viruslike particles obviously undergo lysis. Plaque formation on agar plates could not be seen. It is concluded that the phenomena observed can be compared with cytopathological effects caused by viruses in other cell systems."

X. Laboratory of Medical Microbiology, Dept. of Biology, State University Groningen. Communicated by N. J. W. Kreger-van Rij.

Below follow abstracts of three recent articles.

1. The Structure of Hyphal Septa in Cryptococcus laurentii.

N.J.W. Kreger-van Rij, and M. Veenhuis. 1974. Ann. Microbiol. 24:209.

Summary

Mycelium of Cryptococcus laurentii was studied by electron microscopy. The hyphal septa had central pores of variable width with a well developed or slight swelling of the septum around them.

2. Electron Microscopy of Ascus Formation in the Yeast Debaryomyces hansenii. N.J.W. Kreger-van Rij and M. Veenhuis. Journal of General Microbiology (1975), 89 256-264.

Summary

Ascus formation in Debaryomyces hansenii includes fusion of two cells, usually mother and daughter while still attached to each other, through short protuberances developed from the cross wall between them. Nuclear fusion takes place in the channel connecting the two cells; meiosis apparently occurs in the mother cell. Generally, only one lobe of the meiotic nucleus is surrounded by a prospore wall and it becomes the nucleus of a spore with a warty wall. The rest of the nucleus disappears. The spores germinate by swelling in the ascus and forming one more buds.

3. Conjugation in the Yeast Saccharomycopsis capsularis Schionning. N.J.W. Kreger-van Rij and M. Veenhuis. Arch. Microbiol. 104, 263-269 (1975).

Abstract

Cells of the yeast Saccharomycopsis capsularis fused in pairs after dissolving of part of the cross wall between them near the lateral wall. After nuclear migrations through the opening, the cross wall was closed again and the cells at both sides became asci. The wall of the ascospores developed from a prospore wall. Between the two unit membranes a very thin dark layer broadened to the dark layer of the wall and after that, the light inner layer developed. Immature spores in the strain studied had a ledge which disappeared during maturation.

XI. The Hebrew University, Department of Genetics, Jerusalem, Israel. Communicated by G. Simchen.

Regulation of Mating and Meiosis in Yeast by the Mating-type Region. Y. Kassir and G. Simchen.

A supposed sporulation-deficient mutation of Saccharomyces cerevisiae is found to affect mating in haploids and in diploids, and to be inseparable from the mating-type locus by recombination. The mutation is regarded as a defective a allele and is designated a*. This is confirmed by its dominance relations in diploids, triploids and tetraploids. Tetrad analysis of tetraploids and of their sporulating diploid progeny suggests the existence of an additional locus, RME, which regulates sporulation in yeast strains that can mate. Thus the recessive homozygous constitution rme/rme enables

the diploids a*/α, a/a* and α/α to go through meiosis. Haploids carrying rme show apparent premriotic DNA replication in sporulation conditions. This new regulatory locus is linked to the centromere of the mating-type chromosome, and its two alleles, rme and RME are found among standard laboratory strains.

Effects of the Mitotic Cell-Cycle Mutation cdc4 on Yeast Meiosis. G. Simchen and J. Hirschberg.

The mitotic cell-cycle mutation cdc4 has been reported to block the initiation of nuclear DNA replication and the separation of spindle plaques after their replication. Meiosis in cdc4/cdc4 diploids is normal at the permissive temperature (25°C) and is arrested at the first division (one-nucleus stage) at the restrictive temperature (34°C). Arrested cells show a high degree of recombination commitment (at least 50% of the controls) but no haploidisation. Arrested meiotic cells show a delayed and reduced synthesis of DNA (at most 40% of the control), at least half of which is mitochondrial. It is concluded that recombination commitment does not depend on the completion of nuclear premeiotic DNA replication and that the initiation of this replication might be sufficient for recombination commitment.

Shifts of cdc4/cdc4 diploids to the restrictive temperature at various times of meiosis have revealed three terminal phenotypes, indicating that the function of cdc4 is required for the first division, for the second division and also for spore formation. Thus the primary effect of this gene is not the initiation of DNA synthesis; nor is it likely to be the separation of spindle plaques. It is tentatively suggested that the primary function of cdc4 is related to DNA decondensation.

Recombination and Hydroxyurea Inhibition of DNA Synthesis in Yeast Meiosis. G. Simchen, D. Idar and Y. Kassir.

Hydroxyurea (HU) inhibits the premeiotic DNA replication and the meiotic events that follow, namely readiness, recombination commitment, haploidisation, sporulation commitment and ascus formation. Short incubations with HU (2-4 hrs.) during the premeiotic replication (i.e. starting between 3 and 6.5 hrs in sporulation medium) allow the resumption of the replication at a normal rate following the removal of the drug. The other meiotic events are similarly delayed by the approximate length of the treatment. In these experiments, intragenic recombination in ade2 reached a higher level than in the controls (x1.3-2.0 in one pair of heteroalleles and x3.0-4.0 in another pair). The recombination response to short HU treatments was not observed for a pair of heteroalleles in ade2 that normally shows a high level of meiotic recombination (750 per 10^6 cells) nor was the response observed in a pair of heteroalleles in lys2. HU treatments have almost no effect on sporulating cells from 8 hrs. onwards. At 7-7.5 hrs. the meiotic cells are very sensitive to the drug and even short treatments cause cell death and massive DNA degradation.

Incorporation of ^{3}H-UMP into RNA in Lysing Spheroplasts of Yeast. S. Giami.

Incorporation of radioactive UMP (3H-Uridine - 5'monophosphate) into RNA was examined in spheroplasts of Saccharomyces cerevisiae in a hypotonic

solution. The reaction which takes place in a ρ^- strain, is inhibited by ethidium bromide (250 µg/ml), requires divalent ions, and is only slightly affected by α-amanitin (50 µg/ml). These results are consistent with the idea that the α-amanitin-sensitive RNA polymerase (enzyme II) of yeast is in vivo bound to other factors, thus becoming resistant to α-amanitin under our experimental conditions. Alternatively, enzyme II may be functioning only at a very low rate in this system.

Also published recently: G. Simchen and A. Friedmann: Structure of DNA molecules in yeast meiosis. Nature, 257: 64-66 (1975).

XII. Program in Biology, The University of Texas at Dallas, Richardson, Texas 75080, and Biology Department, University of Dallas, Irving, Texas 75061. Communicated by Herbert Gutz, James H. Meade, and Frank J. Doe.

In the last years, we have been primarily concerned with the genetics of the sexual cycle of Schizosaccharomyces pombe. Most of this work was performed with the Schiz. pombe strains originally introduced into genetic research by Leupold. In these strains, one homothallic (h^{90}), two heterothallic + (h^{+N}, h^{+R}), and one heterothallic - (h^{-S}) mating types were known. The various mating types are determined by two closely linked genes, mat1 and mat2 (h^{90}: mat1$^-$ mat2$^+$; h^{+N}: mat1$^+$ mat2$^+$; h^{+R}: mat1$^+$ mat2^o; h^{-S}: mat1$^-$ mat2^o). In these genotypes, the superscripts + and -indicate alleles of complementary mating activities, whereas the superscript o indicates an inactive allele. For a review of the mating-type system of Schiz. pombe, see references (1) and (2).

Strains of mating type h^{-S} do not mutate to h^{90} or h^+. We found a second h^- mating type, h^{-U}, which mutates spontaneously to h^{90} and h^+. We speculated that h^{-U} may have the genotype mat1^o mat2$^-$ (1). However, recent recombination experiments showed that h^{-U} strains have a mat1$^-$ allele; they may have a new mat2 allele, mat2* (see below).

In the work for his disseration, J. H. Meade performed a detailed analysis of h^+ and h^- mutants isolated from h^{90} strains (7-9). Recombination experiments with two h^+ mutants indicated that their genotype is identical with that of h^{+N} strains. Thus, the mat1$^-$ allele of the h^{90} strain appears to have mutated to mat1$^+$. ——The h^- mutants appear to have mutations in the mating-type gene mat2. Our results show that mat2 not only has a function in copulation and meiosis, but

that it also regulates the formation of the map1 gene product (map1 is a mating-type auxilliary gene). Some of the h^- mutants have lost only one of the three functions while others are defective in at least two, and perhaps all three, functions. All h^- mutants are stable. Thus, their $mat1^-$ alleles, in contrast to those in h^{90} strains, do not seem to mutate to $mat1^+$.

The above results are difficult to explain if, as it is implied in the original two-gene scheme of Leupold, both mating-type genes have similar functions. We found it easier to interpret the results with a modified version of the mating-type scheme of Fincham and Day (Fungal Genetics, Blackwell Scientific Publications, 1963): mat2 is not considered to be a second mating-type gene, but is postulated to be a modifier gene changing $mat1^-$ to a different mating-type specificity. The $mat2^+$ gene has no effect on $mat1^+$. The alleles $mat2^*$ and $mat2^\circ$ are both inactive alleles of mat2, but $mat2^*$ can revert spontaneously to $mat2^+$ whereas $mat2^\circ$ is stable. Furthermore, we propose that $mat2^+$ has three distinct mutator activities which can independely affect the three functions of mat1 (copulation, meiosis, and production of the map1 substance) (8,9).

In other experiments with h^{90} strains, we found mutants which form "mottled" colonies. In contrast to the homogeneously sporulating colonies of h^{90} strains, the mottled colonies consist of sporulating and non-sporulating sectors. When such colonies are restreaked, they yield again mottled colonies. The phenotype "mottled colonies" is caused by mutations in mating type modifier (mmo) genes which are not linked to the mating-type region; four different mmo genes have been found. We postulate that h^{90} cells frequently switch from an h^+ to an h^- activity and vice versa; these switches are assumed to be controlled by the mmo genes (6).

In addition to our work with the Leupold strains, we studied also the mating types of three other Schiz. pombe strains which were isolated in different parts of the world. The strains carry the designations EF4, EF5, and EF6. EF4 and EF5 are homothallic; they form mottled colonies which are similar to those mentioned above. From the EF6

culture, we were able to isolate two heterothallic strains of opposite mating types (h^+ and h^-). In the EF6 strains, mutations from h^+ to h^- and from h^- to h^+ occur rarely; homothallic mutants were not found. Crosses of EF4, EF5 and EF6 with strains of the Leupold material yielded only few viable spores (4). To overcome this disadvantage, we have recently performed a population genetical experiment in which EF6 material was consecutively back crossed with a Leupold strain. We succeeded in the selection of strains which still have the mating-type features of EF6 but which yield a high percentage of viable spores when crossed with Leupold strains.

Dr. James H. Meade has recently assumed a position as research associate with Dr. Thomas R. Manney at the Department of Physics, Kansas State University, Manhattan, Kansas. 66506.

Recent publications and dissertation:

1. Gutz, H., and F. J. Doe (1973): Two different h^- mating types in Schizosaccharomyces pombe. Genetics 74, 563-569.

2. Gutz, H., H. Heslot, U. Leupold, and N. Loprieno (1974): Schizosaccharomyces pombe. pp. 395-446. In; Handbook of Genetics, Vol. 1. Edited by R. C. King, Plenum Press, New York and London.

3. Goldman, S. L., and H. Gutz. (1974): The isolation of mitotic rec^- mutants in Schizosaccharomyces pombe. pp. 317-323. In: Mechanisms in recombination. Edited by R. F. Grell, Plenum Press, New York and London.

4. Gutz, H., and F. J. Doe (1975): on homo- and heterothallism in Schizosaccharomyces pombe. Mycologia 67, 748-759.

5. Meade, J. H., and H. Gutz (1975): A new type of mutation in Schizosaccharomyces pombe: vegetative iodine reaction. Genetics 80, 711-714.

6. Gutz, H., J. H. Meade and Shirley Walker (1975): Mating type modifier genes in Schizosaccharomyces pombe (Abstract). Genetics 80, s38.

7. Meade, J. H., and H. Gutz (1975): Different h^- mutants from homothallic strains of Schizosaccharomyces pombe (Abstract). Genetics 80, s56.

8. Meade, J. H. (1975): Mating-type mutations in Schizosaccharomyces pombe. Dissertation, The University of Texas at Dallas.

9. Meade, J. H., and H. Gutz (1975): Mating-type mutations in Schizosaccharomyces pombe. Submitted to Genetics.

10. Gutz, H., and J. F. Leslie (1975): Gene conversion: a hitherto overlooked parameter in population genetics. Submitted to Genetics.

XIII. Arbeitsgruppe Mikrobengenetik, Fachbereich Biologie, J. W. Goethe-Universitat, 6 Frankfurt/Main, Robert-Mayer-Str. 7-9, Federal Republic of Germany. Communicated by M. Brendel.

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

An improved assay of UV-induced thymine-containing dimers in Saccharomyces cerevisiae. W. W. Fath and M. Brendel. Z. Naturforsch., in press.

Summary

The method for assaying thymine-containing dimers in yeast is based on highly efficient (^{3}H-5'-dTMP) DNA-specific labelling and employs ascending thin layer chromatography. It allows satisfactory quantitative analysis down to UV-doses of 500 erg/mm^2.

UV-induction of thymine-containing dimers in Saccharomyces cerevisiae. W. W. Fath and M. Brendel, Z. Naturforsch., in press.

Summary

In haploid and diploid S. cerevisiae the dimer yield ratio TT/CT is found to be 1.2/1 and 1.3/1, resp., at the UV(254 nm) unit dose 1 erg/mm^2, the share of TT and CT in a UV(254nm) lethal hit being 0.7 TT and 0.6 CT. A general formulation of the UV lethal hit is given and discussed. The TT and CT yields obtained for S. cerevisiae are compared to those reported for other organisms. It is found that there obviously exists a directly proportional linear correlation between genome size and TT + CT yield for the UV dose range well below the saturation levels of the TT and CT formation kinetics.

Nucleic acid metabolism in yeast I. Inhibition of RNA and DNA synthesis by high concentration of exogenous deoxythymidine 5'-monophosphate in 5'-dTMP low-requiring strains. U. G. Langjahr, E.-M. Hartmann and M. Brendel. Molec. gen. Genet., in press.

Summary

The three haploid yeast strains T2tmp1-3, T2tmp1-1, and T6tmp1-51 auxotrophic for 5'-dTMP differ in their requirement for thymidylate: 72, 16 and 3 µg 5'-dTMP/ml will restore optimal growth, respectively. Thymidylate low requirement in strain T2tmp1-1 and T6tmp1-51 is termed tlrA and tlrC, respectively. When the growth medium is made 5 x 10^{-4}M for 5'-dTMP only strain T6tmp1-51 is severely inhibited in RNA and DNA synthesis. This inhibition is reversible after removal of excessive 5'-dTMP. The inhibitory characteristic is in marked contrast to "thymineless death" due to the lack of 5'-dTMP in strain T6tmp1-51 where only DNA synthesis stops while RNA synthesis continues. The inhibitory effect of 5 x 10^{-4}M 5'-dTMP is not due to the 5'-dTMP auxotrophy but to the thymidylate low requiring character (tlrC) in strain T6tmp1-51. The arrest of RNA and DNA synthesis by high concentrations of exogenous 5'-dTMP suggests a regulatory role of either the mono- or triphosphate on nucleoside or nucleotide biosynthesis in yeast.

XIV. Institute of Biochemistry, Section of Molecular Genetics, Bulgarian Academy of Sciences, Sofia 1113, Bulgaria. Communicated by Pencho V. Venkov.

Several years ago we succeeded in the isolation of rare osmotically labile mutants from different wild types of Saccharomyces cerevisiae strains [Biochem. Biophys. Res. Comm. (1974) 53, 599]. These mutants grow exponentially at 30°C in the presence of 10% sorbitol, but are very fragile: when resuspended in buffers they release 50 to 90% of their RNA. Phenotypically, all fragile yeast mutants we have are very similar: they are petite, temperature sensitive at 37°C and show increased sensitivity to a number of antibiotics to which wild types of yeast strains are resistant.

Below follow summaries of our recent studies concerning the action of some antibiotics on RNA synthesis in the fragile yeast mutants.

1. Rifampicin sensitivity of ribonucleic acid synthesis in a fragile S. cerevisiae mutant. P. K. Venkov, G. I. Milchev and A. A. Hadjiolov. (Acc. for publ. in Antimicrobial Agents and Chemotherapy).

RNA synthesis in the sorbitol-dependent, fragile yeast mutant VY1160 is rapidly inhibited by rifampicin. The growth of the mutant cells and protein synthesis are more slowly affected by the antibiotic, apparently as secondary phenomena. Low doses of rifampicin (50 to 100 μg/ml) preferentailly inhibit ribosomal RNA synthesis in comparison to that of messenger and transfer RNA species. Transcription and translation of messenger RNA continues in the presence of low doses of rifampicin as evidenced by the unimpaired induction of α-glucosidase. Partially purified RNA polymerase II from this mutant, in contrast to that from the parental strain is inhibited by low concentrations (1 μg/ml) of rifampicin, while RNA polymerase I from the two strains is similar in behavior. The mutant may be useful for the study of regulatory mechanisms of transcription in eukaryotes.

2. Post-transcriptional degradation of ribosomal and messenger ribonucleic acids in Saccharomyces cerevisiae caused by Actinomycin D. L. W. Watlscheva, P. V. Venkov, B. B. Stoyanova and A. A. Hadjiolov. Biochim. Biophys. Acta (submitted).

The synthesis of RNA in the fragile yeast mutants is inhibited by actinomycin D at concentrations which are without effect on the parental wild type strains. Protein synthesis is not affected initially even at doses which block completely RNA synthesis. Low concentrations of actinomycin D (2 to 5 μg/ml) inhibit preferentially the synthesis of ribonucleic acid as compared to that of poly(A)-containing mRNA and tRNA. Non-specific degradation of prelabelled precursors of ribosomal RNA and poly(A)-containing mRNA is caused by actinomycin D under conditions of complete arrest of new RNA synthesis. This degradation could not be correlated with changes in nuclease activity of yeast cell lysates.

3. Size and turnover of poly(A)-containing messenger RNA in a fragile Saccharomyces cerevisiae mutant. P. K. Venkov, D. Z. Staynov and A. A. Hadjiolov. FEBS Letters (submitted).

Poly(A)-mRNA of S. cerevisiae has been isolated and analyzed under conditions exlcuding degradaition or formation of aggregates. The average molecular weight of total poly(A)-mRNA is 1.2×10^6 d. The half-life of these molecules is 21 min and is idential to that of total yeast mRNA. A correlation is observed between secondary structure and turnover of poly(A)-mRNA molecules.

XV. Ecole Nationale Superieure Agronomique de Montpellier Laboratoire de Recherches de la Chaire de Genetique I.N.R.A. Montpellier - France. Communicated by P. Galzy.

The following articles have appeared recently or will soon be published.

MOULIN, G., RATOMAHENINA, R., GALZY, P. Selection de levure en vue de la culture sur lactoserum. - Le Lait (in press).

Composition in fatty acid, amount of protein and yields on lactose are presented for 27 yeasts able to grow on lactose.

MOULIN, G., RATOMAHENINA, R., GALZY, P., BEZARD, J. Relationship between the presence of linolenic acid and the ability to form respiration deficient mutants in yeast. Folia Microbiologia (in press).

A relationship between the presence of linolenic acid and the ability to form respiration deficient mutants induced by acriflavin has been established. The metabolic coefficient and the composition of fatty acids in samples metabolizing lactose are presented.

BIZEAU, C., BASTIDE, M., GALZY, P., BASTIDE, J. M. JALLAGEAS, J. C. Etude de mutants morphologiques de Saccharomyces cerevisiae HANSEN au moyen de l'immunofluorescence. Ann. Microbiologie, 1975.

The smooth colony mutation in Saccharomyces cerevisiae is under monogenic control. Mutants have lost antigenic sites. Mutants belonging to non-allelic series show differences in their antigenic structures.

BIZEAU, C., JALLAGEAS, J. C., GALZY, P. Utilization of the juice of Helix pomatia for studying the mannans of Saccharomyces cerevisiae HANSEN.

Communication at the fourth international symposium on yeast and other protoplasts. Nottingham, England 8-12th September 1975.

Mannans are prepared by extraction with potassium hydroxyde or by action of snail juice. When studied by magnetic resonnance spectroscopy both preparations show the presnce of α 1-2, α 1-3 and α 1-6 bonds.

Wild strains and mutant "smooth colonies" strains give the same results.

XVI. Department of Genetics, Haryana Agricultural University, Hissar (Haryana) India. Communicated by S. N. Kakar.

Role of recombination in ultraviolet light repair and mutagenesis in Saccharomyces cerevisiae

R. K. Vashishat* and S. N. Kakar

In order to study the role of recombination in repair of radiation damage and damge caused by chemical mutagens, two recombination deficient strains 2C 4 (rec 5) and 2C 8 (rec 4) isolated from the strain Z 140 - 51C were used in these studies. The strains are disomic for chromosome VIII and deficient in X-ray - and ultraviolet light (UV) - induced mitotic heteroallelic reversion. It has been observed that as compared to Z 140 -51C, the strain 2C 4 is more sensitive to UV light, nitrous acid, ethylmethane-sulphonate and N-methyl-N'-nitro-N-nitrosoguanidine. Both the strains are resistant to X-rays at doses between 1.47 to 8.82 kr. The results show that recombination is involved in the repair of UV damage and damage caused by chemical mutagens.

The survival of wild type and of recombination deficient strains is decreased when caffeine (1 g/lit.) is present in the post-irradiation medium. However, there is a significant increase in survival after photoreactivation.

The UV-induced reversion studies on these strains have shown that recombination is also involved in UV-induced mutations because the reversion frequency at ade 2 and his 5 loci is reduced in the 2C 4 strain in comparison to Z 140-51 C.

*present address: Department of Genetics, Punjab Agricultural University, Ludhiana, India.

XVII. Division of Biological Sciences, National Research Council of Canada, Ottawa, Canada K1A OR6. Communicated by M. A. Hannan.

Variation in Radiation Sensitivity During Growth Phase in Mutants of Schizosaccharomyces pombe.

M. A. Hannan and A. Nasim.

A large number of radiation-sensitive mutants have been isolated and characterized in Schizosaccharomyces pombe (Nasim and Smith, 1975, Genetics 79, in press). Several such mutants were biochemically analyzed and it was found that these were capable of excising pyrimidine dimers induced by UV (Birnboim and Nasim, 1975, Mol Gen. Genet. 136, 1-8). It, therefore, seemed worthwile to investigate other possible factors including the absence of a different kind of repair system which might account for the enhanced radiosensitivity of S. pombe mutants.

Eight mutants known to be UV sensitive, were tested for UV sensitivity in different phases of growth, by irradiating cells from log and stationary phase cultures. Itwas observed that all of these mutants yielded survival curves with initial shoulders when cells were irradiated from stationary

phase of growth while such shoulders disappeared completely in survival curves derived from log phase cultures (24 hrs old at 2-5 x 10^6 cells/ml). These results obviously showed a much greater UV sensitivity of the mutants during the log phase of growth. It should be pointed out that the wild type (972 h^-) strain in log phase growth (24 hrs old, 2-5 x 10^6 cells/ml) resulted in survival curves exhibiting slightly greater resistance to UV compared to stationary phase cultures.

The probable factors responsible for enhanced UV sensitivity in the log phase cells of the mutants are being further investigated. Our preliminary findings wiht one mutant (rad-9) indicated that these cells, following UV-irradiation, begin to divide almost immediately in fresh growth medium, thus showing no significant division-delay that is observed in irradiated wild type cells. This would probably mean that the mutant cells divided without giving a chance to any repair system to cope with the UV-induced lesions before they became irrepairable. A class of mutants in which radiation sensitivity could be correlated with the lack of UV-induced division delay would constitute an interesting group of strains for further investigation. Experiments are in progress to determine the nuclear stages of the mutant cells during a growth phase when they are most UV sensitive.

XVIII. Institute of Physics, College of General Education, University of Tokyo, Komaba 3-8-1, Meguroku, Tokyo, 153, Japan. Communicated by Takashi Ito.

Below follow abstracts of three recent papers.

1. T. Ito and K. Kobayashi: Studies on the induction of mitotic gene conversion by ultraviolet irradiation. I. Analysis of dose-frequency relationship. Mutation Research, 30, 33-42 (1975).

Summary

The UV (270-nm) dose-frequency relationship for the induction of intragenic mitotic recombination at trp 5 locus in Saccharomyces cerevisiae was non-linear. Two parameters, α and a, in the proposed equation for the non-linear relationship $f = (at)^{\alpha}$ were determined so as to fit the experimental data by the method of least squares. The analysis was extended over 5 cell stages during synchronous growth. It was found that (1) parameter α changed from 2.02 for unbudded small cells to 1.09 for the stage where the cell had finished the division of the nucleus, and (2) parameter a changed correspondingly from $7.25 \cdot 10^{-4}$ to $0.180 \cdot 10^{-4}$ sec^{-1} during the same period.

One interesting outcome in this analysis was the deduction of a dose-dependent nature of relative sensitivity with respect to the stage. The determination of these two parameters enabled us to calculate dose-effect relationships beyond the limits of experimental restrictions. Such an "imaginary" relationship, calculated at an extremely low dose, revealed the existence of maximal sensitivity around the DNA synthesis period. It was further shown that this maximum would easily be masked even in the moderate dose range. Thus, we conclude that the validity of single dose comparisons is diminished unless α is constant regardless of the cell stage. Some considerations on the proposed parameters have been made in relation to the mechanisms of the induction of gene conversion by UV.

2. T. Ito and K. Kobayashi: Studies on the induction of mitotic gene conversion by ultraviolet irradiation. II. Action spectra. Mutation Research, 30, 43-54 (1975).

Summary

Action spectra for the induction of intragenic mitotic recombination (gene conversion) at the trp 5 locus by UV are presented for three cell stages (T_0, T_9 and T_{16}) taken from synchronously growing cultures of Saccharomyces cerevisiae. The spectra over the range from 230 to 300 nm were taken mostly in 5-nm steps. The peak of action spectra was significantly shifted, regardless of the stage, toward the longer wavelengths as compared with that of the absorption spectrum of DNA (258 nm) or even that of thymine (265 nm). In one extreme case (T_{16}), the peak was shifted 17 nm from the absorption peak of DNA. Further, the spectrum changed its shape as the cell stage advanced from non-dividing (unbudded) (T_0) to a dividing phase (T_{16}). Furthermore, the induction cross section decreased by a large factor (about 40), regardless of the wavelength, in going from T_0 to T_{16}. From observations of the high photoreversibility of induced conversions, the major primary damage was thought to be pyrimidine dimers in the DNA.

One plausible explanation, though not quite satisfactory from the quantitative viewpoint, for these findings was that the increasing RNA during growth would screen the incident UV differentially with respect to the stage. If this explanation is correct, thymine dimers may still be considered, in spite of the shifts and deformations in the action spectra, as the major primary damage that triggers the long series of processes leading to gene conversion. Conventional methods for obtaining action spectra are discussed in comparison with the present method, which was based on sensitivity parameter a in the proposed dose (t)-frequency (f) relation, $f = (at)^{\alpha}$ (α is the multiplicity parameter).

3. K. Kobayashi and T. Ito: Further in vivo studies on the participation of singlet excited oxygen as the intermediate in the photodynamic inactivation and induction of genetic changes in Saccharomyces cerevisiae. Photochemistry and Photobiology (in the press) (1975).

Summary

In vivo participation of singlet excited oxygen in the photodynamic inactivation and induction of genetic changes (gene conversion) in acridine orange-sensitized yeast cells was investigated by using N_3^-, an efficient singlet oxygen quencher, and D_2O, a known agent for the enhancement of the life-time of singlet oxygen. The addition of N_3^- protected the cells from both photodynamic actions. From an anlysis of the concentration-dependent protection, about 80% of the induction of the genetic change is explainable on the basis of singlet oxygen mechanism. The quantitative estimation of the N_3^- protection in the inactiviation was not possible because of the sigmoidal nature of the inactivation curve. The replacement of H_2O with D_2O during illumination was effective in enhancing the photodynamic inactivation but almost completely ineffective for the gene conversion induction. The deuterium effect with the cell system was clearly not as large as would be expected from in vitro experiments. This, however, could be explained

from the kinetic consideration that natural quenchers of singlet oxygen in the cell would mask the deuterium effect. By experiments with different cell stages it was demonstrated that these two modifying effects were dependent on the intracellular reaction environment. The conclusion is that singlet oxygen must be the major intermediate responsible for the photodynamic actions in acridine orange-sensitized yeast cells.

XIX. University of Nancy, Biochimie Microbienne, 5, rue Albert Lebrun, 54001 Nancy Cedex, France. Communicated by R. Bonaly.

During last year, studies on Rhodotorula yeast cell-walls have been carried out. The investigations were concerned with the chemical structure, enzyme composition and the influence of antibiotics and antifungal substances on the yeast-walls.

The results have been published in the following reviews:

Incidence du traitement par l'ethylene diamine sur l'ultrastructure des parois de trois levures du genre Rhodotorula. (1975) C. R. Acad. Sc. Paris 280, 1417-1420.

Etude des parois de levures Rhodotorula.

V. Influence des conditions de culture sur les phosphatases de Rh. rubra (1975) Biochim, Biophys. Acta 392 39-50.

VI. Influence du 2 hydroxybiphenyle sur les activites phosphotasiques de Rh. rubra. (1975) Biochim. Biophys. Acta 392 51-63.

Modifications chimiques et ultrastructurales de la paroi de Rh. rubra cultivee en presence de chloramphenicol ou de 2 hydroxybiphenyls (1975) C. R. Acad. Sc. Paris 280 1169-1172.

Etude des parois de Rhodotorula rubra: Influence du chloramphenicol sur la morphologie et les phosphatases des parois de Rh. rubra. Chemico. Biological Interactions: in press.

We will carry on this year our studies on the chemical structure of the cell walls with a particular interest in the chemical modifications induced by the antifungal substances when present in the growth medium of the yeasts.

XX. University of Salamanca, Department of Microbiology, Salamanca, Spain. Communicated by V. Notario, J. R. Villanueva and T. C. Villa.

Research on Yeast Hydrolases.

Recently we have made a revision concerning the distribution of β-glucanases and their possible functions in nature (1). The main roles that such enzymes carry out in nature can be ascribed to metabolic, morphogenetic lytic and ecological aspects.

We have also considered the biochemical properties of yeast and fungal β-glucanases. The principal function of these enzymes in yeasts is related to the processes of cell wall growth and budding.

β-(1→3)-glucanases in several yeasts have been the main subject of the investigations in our laboratory. Recent studies report the results on the β-(1→3)-glucanases of Pichia polymorpha (2) and Cryptococcus albidus var. aerius. β-(1-3) glucanase activities have been found in cell-free extracts as well as in the culture medium of these yeasts. The maximum production of β-glucanases in asynchronous cultures of P. polymorpha and Cr. albidus var. aerius takes place in log-phase cells where about 80% of the cells are budding.

Mild acid treatment of whole yeast cells was carried out to determine the cellular location of β-glucanase activities present in cell-free extracts of both yeasts. These experiments have shown that β-glucanases are mainly located outside the plasma membrane and not covalently linked to the cell wall. However, in Cr. albidus var. aerius 50% of the β-glucanase activity was slightly bound to the cell wall.

A recent study reports the identification of three different enzymes in cell-free extracts as well as in the culture medium of P. polymorpha that hydrolyze laminarin and only one that hydrolyses pustulan. The three β-glucanases appear to be three different enzymes. Evidence for different molecular weights, Km values and patterns of activity has also been obtained. These same three molecular forms have been detected in the medium for growing protoplasts. In every case these forms have been purified and characterized.

Sephadex G-50 filtration on culture samples obtained at different times during the growth of Cr. albidus var. aerius have shown the existence of several molecular forms of β-glucanase in cell-free extracts and in the culture medium. All of these forms present activity against β-(1-3)- and β-(1-6)-glucans. These results and those obtained by treatment of the different samples with Triton X-100 and EDTA at room temperature suggest the possibility either of replacement changes in fractions containing β-glucanase activity or of a different synthesis of each β-glucanase during the growth of the yeast.

The lowest molecular weight form of β-glucanase was isolated and purified from cell-free extracts. This low-molecular weight form may may be considered as a non-specific exo-β-glucanase. The Km values were calculated with purified enzyme acting on p-nitrophenyl-β-D-glucopyranoside, pustulan and laminarin. An inhibitory effect on the action of this enzyme was observed at high concentrations of pustulan and laminarin. The extent of inhibition was highest in the presence of pustulan. Such an inhibitory effect was not found when p-nitrophenyl-β-D-glucopyranoside was used as substrate. Molecular weight, determined on a Bio-gel P-10 column was estimated to be ± 2100 daltons.

Studies of the effect of 2-deoxy-D-glucose on the synthesis of β-glucanase, invertase and acid phosphatase by cells and protoplasts of the yeast P. polymorpha have been carried out in our laboratory (4). Our results indicate that the synthesis of β-glucanase either by cells or by protoplasts occurs in the presence of the glucose analogue in the culture medium. On the other hand, the synthesis of the other typical extracellular proteins is strongly affected by the drug. The

degree of inhibition depends on the 2-deoxy-D-glucose concentration. Inhibition of cell wall resynthesis and the action of β-glucanase would produce lysis of yeast cell walls when the cells were grown on 2-deoxy-D-glucose.

More recently (5) we have reported the presence of β-xylosidase activity in cell-free extracts and in the culture medium when Cr. albidus var. aerius was grown on glucose as the sole carbon source, appearing to be a constitutive enzyme. Mild acid treatment of whole cells suggests that the total intracellular activity is located in the periplasmic space and some experiments indicate that it is partially associated with the cell wall. This enzyme has been partially purified by Sephadex gel filtration and DEAE chromatography. DEAE Sephadex A_{50} chromatography has shown that there are two different forms of β-xylosidase in cell-free extracts whereas only one form is present in culture fluids.

References

1. Villanueva, J. R., Notario, V., Santos, T., Villa, T. G. (1975) 4th International Symposium on Yeast and other Protoplasts. Academic Press (in press).

2. Villa, T. G., Notario, V., Villanueva, J. R. (1975). Archiv. Microbiol. 104, 201-206.

3. Notario, V., Villa, T. G., Benitez, T., Villanueva, J. R. (1975) Can. J. Microbiol. (in press).

4. Villa, T. G., Notario, V., Benitez, T. Villanueva, J. R. (1975) Archiv. Microbiol. (in press).

5. Notario, V., Villa, T. G., Villanueva, J. R. (1975). Can. J. Microbiol. (in press).

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

1. H. J. Phaff delivered the Annual Lecture before the Mycological Society of America in Corvallis, Oregon on August 21, 1975. His topic was "Speciation in Yeast - an Approach by Genome Comparison."

2. Dr. Melvin T. Meyer completed his Ph.D. dissertation in August 1975 under the guidance of Professor H. J. Phaff. Dr. Meyer is now doing postdoctoral research in the laboratory of Professor D. J. Manners, Heriot-Watt University, Edinburgh, Scotland. He expects to remain there until September 1976. The following is an abstract of the dissertation, entitled α-(1→3)-Glucanases from Bacillus circulans WL-12 and from Certain Yeasts.

Endo-α-(1→3)-glucanase is produced in high concentration in the culture fluid of Bacillus circulans WL-12 when grown in a mineral medium on Schizosaccharomyces pombe whole cells or on extensively purified α-(1→3)-glucan as the sole carbon source. Sch. pombe cell walls or a crude α-(1→3)-glucan extract resulted in higher levels of β-(1→3)- and

β-(1→6) glucanases than with highly purified β-(1→3)-glucan or whole cells.

The endo-α-(1→3)-glucanase was separated from the bulk of the β-glucanases by affinity adsorption of the enzyme on insoluble α-(1→3)-glucan, followed by the release of the enzyme by subsequent autohydrolysis of this substrate. The enzyme was purified further by DEAE-agarose chromatography and polyacrylamide P-150 gel filtration. The first step, passage over DEAE-agarose, also separated a minor α-(1→3)-glucanase component. The principal endo-α-(1→3)-glucanase is specific for the α-(1→3)-glucosidic bond of α-(1→3)-glucan (pseudonigeran), and of carboxymethyl-α-(1→3)-glucan, but it does not appeal to hydrolyze nigerobiose. The enzyme conforms to Michaelis-Menten kinetics with pH optimum 7.5 to 8. The hydrolytic reaction does not proceed at a constant rate with either α-(1→3)-glucan as substrates. The enzyme has no metal ion requirement but does contain essential sulfhydryl groups. Several divalent heavy metal ions are inhibitory. The molecular weight by SDS-polyacrylamide gel electrophoresis, was estimated to be 135,000 daltons ± 10% with no evidence for subunit structure.

Several selected yeast species, known or suspected to contain α-(1→3)-glucan in their cell walls, were screened for the presence of an endogenous α-(1→3)-glucanase. Carboxymethyl-α-(1→3)-glucan was used as the assay substrate to increase detection sensitivity. Three Schizosaccharomyces species and Phaffia rhodozyma, all of which contain α-(1→3)-glucan in the cell wall, were negative for α-(1→3)-glucanase activity. Cryptococcus species, which also contained cell wall α-(1→3)-glucan, exhibited α-(1→3)-glucanase activity in their cell walls and intracellular extracts. α-(1→3)-glucanase was also detected in Rhodotorula species, although α-(1→3)-glucan has not been demonstrated in species of this genus.

The α-(1→3)-glucanase in Rhodotorula minuta var. texensis was studied in limited detail. The cell wall and intracellular enzyme activities were very unstable when stored for 24 h at 4 C. Dialysis of the fresh intracellular extract activated an inactive fraction of the enzyme. The activated portion of the α-(1→3)-glucanase was also susceptible to inactivation. Storage of the fresh extract for several days, followed by dialysis led to activity, indicating that the inactive form in the fresh extract is protected to some extent by avoiding dialysis. EDTA also led to considerable inactivation, suggesting an essential metal ion requirement for activity.

3. Below follow abstracts of current papers from our laboratory.

Miller, M. W., M. Yoneyama and M. Soneda. Phaffia, a new yeast genus in the Deuteromycotina (Blastomycetes). Internat. J. Syst. Bacteriol. (accepted for publication).

A description is given of a new yeast genus Phaffia, represented by P. rhodozyma sp. nov. to accommodate nine yeast strains isolated in Japan and one in Alaska, U.S.A., all from exudates of deciduous trees. The type strain of P. rhodozyma is UCD (FS&T) 67-210 (= CBS 5905). Phaffia, named in recognition of the contributions of H. J. Phaff to yeast taxonomy and ecology, is a carotenoid-producing, fermentative yeast of the Deuteromycotina (Blastomycetes) whose properties indicate a basidiomycetous origin. A comparision is made between Phaffia and other yeast genera to which it might be related.

Torulopsis sonorensis, a new species of the genus Torulopsis. M. W. Miller, H. J. Phaff, Mary Miranda, W. B. Heed and W. T. Starmer. Internat. J. Syst. Bacteriol. (accepted for publication).

A novel member of the yeast genus Torulopsis has been recovered 35 times during 1971, 1972, 1973, and 1974 from Drosophila mojavensis, from soft-rot pockets from six species of cacti, and from soil wetted by soft-rot fluid. The collections were made in the Sonoran desert of southern Mexico and of northern Mexico. The new species was named Torulopsis sonorensis after the geography of its habitat. The type strain is UCD (FS&T) 71-148 (= ATCC 32108 = CBS 6792).

Fleet, G. H. and H. J. PHaff. Glucanases in Schizosaccharomyces: isolation and properties of exo-β-glucanase from the cell extracts and culture of Schizosaccharomyces versatilis. Biochim. Biophys. Acta (in press).

SUMMARY

(1) Cell extracts and extracellular culture fluids of species of the yeast genus Schizosaccharomyces exhibited exo-β-(1→3)- and exo-β-(1→6)-glucanase activities.

(2) Using a combination of Sephadex G-100 and DEAE cellulose chromatography, the exo-β-(1→3)-glucanases from the cell extracts and culture fluid of Schizosaccharomyces japonicus var. versatilis were extensively purified. The enzymes from either location exhibited similar purification and other properties.

(3) The purified enzymes hydrolyzed the β-(1→6)-glucosidic linkage in addition to the β-(1→3)-linkage. Heat denaturation, inhibition, and electrophoretic studies indicated that both hydrolytic activities were the properties of a single protein. Laminarin and pustulan hydrolysis followed Michaelis-Menten kinetics. The Km and V for laminarin hydrolysis were 6.25 mg/ml and 350 μ moles of glucose released/min/mg protein, and for pustulan they were 166 mg/ml and 52 μ moles of glucose released/min/mg protein.

(4) The exo-β-glucanase was assigned a molecular weight of 43,000.

(5) The purified enzyme failed to hydrolyze isolated cell walls from either baker's yeast or Schiz. pombe or to induce protoplast formation from intact cells of Schiz. versatilis or Saccharomyces cerevisiae.

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Fleet, G. H. and H. J. Phaff. 1974. Glucanases in Schizosaccharomyces. Isolation and properties of the cell wall-associated β-(1→3)-glucanases. J. Biol. Chem. 249, 1717-1728.

Mendonca-Hagler, L. C. and H. J. Phaff. 1975. DNA base composition and DNA/DNA hybrid formation in psychrophobic and related yeasts. Internat. J. Syst. Bacteriol. 25, 222-229.

XXII. The Finnish State Alcohol Monopoly, Alko, Box 350, SF-00101, Helsinki 10, Finland. Communicated by Heikki Suomalainen.

On the Activity and Regulation of Anaplerotic and Gluconeogenetic Enzymes During the Growth Process of Baker's Yeast. S. Haarasilta and E. Oura. Eur. J. Biochem. 52: 1, 1-7, 1975.

The anaplerotic and glucongeogenetic metabolism of baker's yeast growing in a chemostat with oxygen and/or carbon source as the growth limiting factor was studied at the enzymatic level. Under anaerobic conditions on glucose the pyruvate carboxylase reaction was the only active anaplerotic mechanism. Its activity was greatly reduced when yeast was grown on glucose in the presence of aspartate. The glyoxylate cycle and the specific enzymes of gluconeogenesis, phosphoenolpyruvate carboxykinase and hexosediphosphatase, were completely repressed under anaerobic conditions. Their catabolite repression could be partially reversed using quite high oxygen tensions or by using glycerol, pyruvate or ethanol as carbon sources in place of glucose or galactose. In a non-respiratory variant of Saccharomyces-yeast they were completely repressed independently of aeration conditions.

Soluble and Membrane-bound Cyclic AMP Diesterase Activity with a Low Michaelis Constant in Baker's Yeast. J. C. Londesborough. FEBS Letters 50:2, 283-287, 1975.

The total 3', 5'-cyclic AMP (cAMP) diesterase activity of baker's yeast at 0.2 μ M cAMP is roughly equal to the adenyl cyclase activity (6 n mole/min/g/ fresh yeast). The high Km, 65 000 mol. wt. soluble cAMP diesterase (described earlier), accounts for only about 13% of this total. The rest is due to a 140 000 mol. wt. soluble enzyme and a particle-bound enzyme. Both have Michaelis constants of about 0.15 μ M, and in contrast to the high Km enzyme, require divalent metals and are not inhibited by thiols. The particle-bound activity is reversibly eluted from the 105 000 g precipitate by 0.3 M KCl.

CO_2-Fixation, Theoretical Background. E. Oura. Proc. 4 Int. Symp. Yeasts, Vienna, Austria 1974, Part II, pp. 58-59.

Some aspects of the CO_2-fixation process in yeast, particularly in baker's yeast grown on glucose or ethanol are examined on a theoretical basis. The only significant reactions by which yeast assimilates CO_2 are (a) the formation of catbamyol phosphate, (b) the assimilation of CO_2 to form P-ribosyl-aminoimidazole-carboxylic acid, and (c) pyruvate carboxy-lase (PC) reaction. The first two reactions are active during all growth conditions covered by this statement. The PC reaction (a) functions as the only OAA regenerating reaction in yeast growing anaerobically on glucose, (b) in yeast growing aerobically on glucose this reaction can be fully or partially replaced by the glyoxylate cycle, which functions independently of CO_2-fixation, and (c) PC reaction is unnecessary in yeast growth on ethanol.

It is possible to calculate that when OAA regeneration proceeds *via* PC reaction ca. 6.3% of yeast cell carbon is derived from assimilated CO_2 whereas when the glyoxylate cycle fully replaces the PC reaction, the corresponding value is only 1.3%. In our experiments we have, in fact, obtained smaller values for CO_2-fixation during growth on ethanol than during growth on glucose.

Although yeast fixes CO_2 e.g. in the reaction where malonyl-CoA is formed from the corresponding acetyl compound, the fixed C-atom is split off in the following step of the reaction sequence in fatty acid synthesis. Some normally decarboxylative reactions are in *vitro*, and perhaps under unusual conditions also in *vivo*, capable of reversal. There is, however, no possibility of permanent fixation of considerable amounts of CO_2 into cellular material *via* these reversed reactions. It is apparent that yeast of normal cell composition is capable of gaining a maximum of 6-8% of its cellular carbon from assimilated CO_2. If higher values are obtained they may perhaps be explained by (a) unusually high concentrations of nucleic acids and amino acids from the aspartate and glutamate families in the yeast strain used, (b) by as yet unknown CO_2-fixation reactions in yeast, or (c) by incorrect experimental design or interpretation of results. Using a definite quite small CO_2 tension during growth, yields up to 25% greater than those normally considered maximal have been obtained. As this increase can hardly be explained by an increase in CO_2-fixation an explanation must be sought elsewhere. Some hypotheses can be formed on the basis that CO_2 either influences directly or indirectly the metabolism on enzymatical or, preferably, energetical level.

On the Formation of Glycogen and Trehalose in Baker's Yeast. S. Grba, E. Oura and H. Suomalainen. (Submitted for publication in Eur. J. Appl. Microbiol.).

More glycogen and trehalose is formed in aerobically incubated baker's yeast than under anaerobic conditions, glucose being a more favorable source of sugar than maltose. The regulation of the formation of glycogen in aerobic incubations of *non-proliferating* baker's yeast in the presence of glucose can be explained by the action of activators and inactivators. The level of ATP in the cell does not affect the formation of trehalose in the same way as it influences the formation of glycogen.

The incubation temperature chosen can be used to manipulate the relative proportions of glycogen and trehalose in baker's yeast. 30°C is the optimum for the formation of glycogen, and at 45°C none at all is formed. The inhibition of the biosynthesis of glycogen is not, at least primarily, a consequence of the effect of the elevated temperature on the enzymes taking part in the formation of glycogen. The optimum temperature for the formation of trehalose is 45°C, and at this temperature baker's yeast containing as much as 20% trehalose can be obtained.

Effect of Aeration on the Activity of Gluconeogenic Enzymes in Saccharomyces cerevisiae Growing under Glucose Limitation. S. Haarasilta and E. Oura. (Submitted for publication in Archives of Microbiology).

The effect of aeration intensity on the levels of the key enzymes of gluconeogenesis (phosphoenolypyruvate carboxylase) was studied in baker's yeast (Saccharomyces cerevisiae) and in a non-respiratory variant of Saccharomyces cerevisiae grown in a chemostat under glucose limitation.

In baker's yeast phosphoenolpyruvate carboxykinase, hexosediphosphatase and isocitrate lyase were completely repressed under anaerobic conditions. Their repression, especially that of hexosediphosphatase, could be partially reversed by using intense aeration. In the non-respiratory variant these enzymes were completely repressed independently of aeration. Pyruvate carboxylase of baker's yeast showed maximal activity under anaerobic conditions, the activity decreasing rapidly with only slight aeration. In the non-respiratory variant pyruvate carboxylase had low activity under both anaerobic and aerobic conditions.

Quantitative Estimation of 3'5'Cyclic AMP Phosphodiesterase Using Anion Exchange Resin in a Batch Process. J. Londesborough. (Subm. for publication in Anal. Biochem.)

A rapid and quantitiative assay system for cyclic AMP phosphodiesterase is described. The method is based on the snake venom procedure of Thompson and Appleman (Biochemistry 10 (1971), 311-316), but the recovery of adenosine is increased from 60% to 100% by using Dowex anion exchange resin suspended in a mixture of ethanol and water.

With this assay procedure, and either crude microsomes or whole microsomal supernatant of baker's yeast source, the loss of 3H-cyclic AMP (initially 0.25 μ M) equals the gain of 3H-adenosine recovered in Dowex supernatants.

On the Enzymic Composition of Yeast Nuclei and Their Membranes. J. Londesborough and K. Varimo. (Abstr. of paper presented at the 4th Intern. Symp. on Yeast and Other Protoplasts, September 8-12, 1975, Nottingham).

Nuclei with a DNA:RNA:Protein ratio of 1:5:20 were isolated from semi-aerobic baker's yeast. They contained only 0.5% of the total hexokinase activity and less than 10% each of the adenyl cyclase and Nicotinamidemononucleotide Adenyl Transferase ("NAT") activities. Adenyl cyclase was extensively solubilized by the buffers used, but this artefact does not account for the small NAT activity in yeast nuclei compared to rat liver nuclei. The isolated yeast nuclei contained about 40% and 20%, respectively, of NADPH- and Succinate cytochrome c reductase activities, probably indicating that the former enzyme, at least, is specifically concentrated in the nuclear membrane.

Behaviour of Plasma Membrane Fragments During Zonal Centrifugations of Homogenates from Glucose-Repressed Saccharomyces cerevisiae. T. Nurmine, L. Taskinen and H. Suomalainen. (Submitted for publication in The Biochemical Journal).

1. The distributions of several enzymes and other marker components were examined after zonal centrifugations of whole homogenates from glucose-repressed Saccharomyces cerevisiae on sucrose and iso-osmotic Ficoll, and the composition and morphology of the fractions were investigated. 2. After high-speed zonal centrifugation, most of the protein, acid and alkaline phosphatase, alkaline pyro-phosphatase, adenosine monophosphatase, β-fructofuranosidase, α-mannosidase, NADPH-cytochrome c oxidoreductase and an appreciable amount of phospholipid and sterol were non-sedimentable, i.e. were at densities below 1.09. Most of the RNA was at 1.06-1.08 ρ in Ficoll and at 1.09-1.11 ρ in sucrose. 3. The bulk of the MG^{2+} - dependent adenosine triphosphatase (Mg-ATPase) was coincident with the main peak of phospholipid and sterol, at median density 1.10, which was also rich in light vesicles of plasma membrane. In Ficoll, a minor peak of phospholipid and sterol at 1.12-1.15 ρ contained a smaller part of the ologomycin-insensitive Mg-ATPase and heavy fragments of plasma membrane. In sucrose, several minor peaks of Mg-ATPase were in the mitochondrial density range and a peak of oligomycin-insensitive Mg-ATPase coincident with a minor peak of phospholipid and sterol at around 1.25 ρ contained heavy fragments of plasma membrane of much higher carbohydrate content, especially mannose, than did the lighter vesicles of plasma membrane. 4. Further purification of the plasma membrane preparations was achieved on Urografin gradients. 5. It is concluded that the behaviour of the plasma membrane fragments during density gradient centrifugations largely depends on the amount of glycoprotein particles present.

Enthalpy Changes of Yeast Growth Under Different Intensities of Aeration. E. Oura. Fifth Intern. Biophy. Congress of the Intern. Union for Pure and Appl. Biophys., Copenhagen 1975. Abstracts P-456.

The growth process must be treated as a whole when examining the formation of heat during yeast growth; it cannot be separated into an energy producing catabolism and an energy consuming anabolism. The production of heat during the growth process has been estimated from data obtained in glucose-limited continuous cultivations with different intensities of o aeration. Using the calorific content 515 kcal/100 g yeast cell material (d.w.), the heat produced during anaerobic growth is calculated to be about 19 kcal/mole glucose used. This value increases with increasing aeration intensity to become 220 kcal/mole glucose used in fully oxidative growth. The ratio of these values is 1/11. On the other hand the ratio of ATP production during fermentative to that during oxidative metabolism (P/O=2) is 1/14. It is concluded that there is either an energy wastage during aerobic growth or an increased energy requirement during aerobic growth that reflects the higher structural and functional complexity of aerobic yeast cells.

The Effects of the Intensity of Aeration on the Biochemical Composition of Baker's Yeast. III. Activities of Enzymes of the Glycolytic and Pentose Phosphate Pathways. E. Oura. Subm. for publ. in Biotechnol. and Bioeng.

A series of baker's yeast continuous cultivations was made using different intensities of aeration. The experimental conditions were such as to eliminate the effects of catabolite repression on the formation of enzymes. The variation in activity of several enzymes was investigated and distinct changes were noted. The activities of hexokinase and alcohol dehydrogenase characterize the actual fermentation activity of yeast, the same being true, in part, for pyruvate decarboxylase. The activty of phosphofructokinase is nearly insensitive to the oxygen level at normal tensions. The activity of the cell to the phosphofructokinase can be limited in anerobic conditions by its scarcity. The insensitivity of glucose-6-phosphate dehydrogenase to the oxygen tension together with its low activity suggests that this enzyme plays primarily a biosynthetic role and that the function of the pentose phosphate pathway as an energy-producing route is negligible.

The Role of Oxaloacetate as Feed-Back Inhibitor of Isocitratelyase in Baker's Yeast. M. Varimo and E. Oura. (Subm. for publ. in Acta Chem. Scand. B.)

The Michaelis constant for threo-D_s-isocitrate of isocitratelyase from baker's yeast was estimated to be 0.35 mM and the oxaloacetate inhibition constant 1.1. mM. Considering the oxaloacetate concentration in the cells, it could be concluded that oxaloacetate cannot act as a feed-back inhibitor of isocitratelyase in baker's yeast.

The following publications have appeared since the last communcations. The abstracts of reports have been given in Yeast News Letter 23:1, 14, 17, 1974 and 23:2. 71, 1974.

Oura, E., Effect of aeration intensity on the biochemical composition of baker's yeast. I. Factors affecting the type of metabolism. Biotechnol. Bioeng. 16, 1197-1212, 1974.

Oura, E., Effect of aeration intensity on the biochemical composition of baker's yeast. II. Activities of the oxidative on enzymes. Biotechnol. Bioeng. 16, 1213-1225, 1974.

Suomalainen, H., Some enzymological factors influencing the leavening capacity and keeping quality of baker's yeast. Eur. J. Appl. Microbiol. 1, 1-12, 1975.

Suomalainen, H. & Nykanen, L. and Eriksson, K., Composition and consumption of alcoholic beverages. - A review. Amer. J. Enol. Viticult. 25, 179-187, 1974.

Nurminen, T. & Konttinen, K. and suomalainen, H., Neutral lipids in the cells and cell fractions of aerobic baker's yeast and anaerobic brewer's yeast. Chem. Phys. Lipids 14:1, 15-32, 1975.

XXIII. Oregon State University, Department of Microbiology, Corvallis, Oregon, 97331. Communicated by L. W. Parks.

The following is an abstract of some recent work by R. B. Bailey and L. W. Parks.

Variation in the percentage of sterols esterified to long chain fatty acids during cellular growth has been examined. Under all conditions a constant percentage of sterol esters was maintained during exponential growth. However, this maintenance level varied with different carbon sources and appeared to be proportional to the growth rate. A sharp increase in the rate of esterification was observed upon entry of the culture into the stationary growth phase. The minor cellular sterol components were found to accumulate following this period of rapid sterol ester synthesis, with a relative decrease in the size of the ergosterol pool. Sterol esters of ergosterol precursors are unable to be metabolized to ergosterol. Once esterified, the fatty acids don't appear to be scavenged during starvation conditions.

XXIV. Mikrobiologisches Institut, Swiss Federal Institute of Technology, Weinbergstrasse 38, CH-8006, Zurich, Switzerland. Communicated by A. Fiechter.

Recent Publications

1. A. Einsele, H. Schneider, and A. Fiechter: Characterization of Microemulsions in a Hydrocarbon Fermentation by Electronmicroscopy. J. Ferm. Technol. 53 (4), 242, 1975.

It has been experimentally demonstrated that a microemulsion is built up in fermentation of n-hexadecane with Candida tropicalis. These submicroscopic droplets (< 1 μm) are characterized by electron-microscopy.

2. O. Kappeli, H. Aeschbach, H. Schneider, A. Fiechter: A Comparative Study of Carbon Energy reserve metabolism of Candida tropicalis growing on glucose and on hydrocarbon. Europ. J. Appl. Microbiol. (in press).

Glycogen was markedly accumulated in C. tropicalis growing on glucose with increasing limitation of external substrate supply. The same effect was caused by a N-free medium. The lipid content did not show any significant change in both cases. On the hydrocarbon substrate lipid increased as substrate availability decreased, whereas glycogen accumulation was only slight. However, the increase of lipid content on hydrocarbons did not reach the same level of accumulation as glycogen on glucose. In N-free medium both glycogen and lipids accumulated, indicating that glycogen is not substituted by lipids as the carbon energy reserve on a hydrocarbon substrate.

In addition a refined shift-technique is described. The disturbing influence of excess substrate at the beginning of a shift from glucose to hydrocarbons is avoided by a portioned substrate feeding according to the cell activity.

3. A. Zimmerli: Zur Lokalisation der Malatdehydrogenase in Schizosaccharomyces pombe. Swiss Federal Institute of Technology, Disseration Nr. 5469, Zurich, 1975.

Four forms of malatedehydrogenase can be shown in Schizosaccharomyces pombe. The enzyme has been localized by separating mitochondria from the cytoplasm.

Identification by the isoelectric point (pI) of the malate dehydrogenase in disintegrated mitochondria and in a mitochondrial-free extract showed the presence of two different enzyme forms. One form with an isoelectric point of 6.3 is localized in mitochondria, another form with an isoelectric point of 5.7 in the cytoplasm.

The performance of the zonal rotor has been improved by the construction of a new zeal assembly.

4. H. Hug, H. W. Blanch, and A. Fiechter: The Functional Role of Lipids in Hydrocarbon Assimilation. Biotech. Bioeng. 16, 965-985, 1974.

The yeast Candida tropicalis utilizes both glucose and hydrocarbons as sole carbon sources. When grown on hydrocarbons, the cells contain twice as much lipid as when grown on glucose. In transient continuous culture experiments, following a substrate change from glucose to hexadecane, an adaption phase occurred. During this phase the lipid concentration per cell increased greatly. It is proposed that a high cellular lipid concentration is necessary for hydrocarbon assimilation, and this is not just a reflection of the lipophilic nature of the substrate.

5. A. Fiechter: Regulatory aspects of Yeast Metabolism and their consequences for cell mass production. Proc. 4th Int. Symp. Yeasts, Vienna, Part II p. 17-33, 1974.

High productivity is a decisive prerequisite for attainment of economy in single cell production. - Mass transfer rates for transport of substrate and oxygen at permissive costs for power input is still critical. - Regulatory effects of glucose and oxygen might lower the biosynthetic potential. Proper choice of the external reaction parameters are mandatory for maximum output. - At present we are not able to ascertain the exploitation of the true biosynthetic potential of the cell.

6. H. W. BLanch, A. Fiechter: Dispersion and Coalescence Phenomena in the Hydrocarbon Fermentation.

The average times between coalescences of droplets in fermentation conditions reported here are an order of magnitude greater than in pure systems and at lower levels of agitation are sufficient such that one droplet may be completely consumed by a single cell growing at a specific growth rate of 0.2 to 0.3 hr^{-1}. In chemostat operation with pure substrate, the coalescence frequencies will be critical, especially where segregation effects become important. Some of the models proposed must be thus reassesed in this light. If the substrate is dissolved in an inert dispersed phase, concentration differences will result

from drop to drop due to the long times between coalescences and droplet size distribution. In designing fermenters for hydrocarbons it may thus be necessary to provide a region of intimate contacting of drops and cells; the previous results of continuous cultures may reflect the effects of incomplete mixing and the long times between coalescences.

XXV. Research Institute of Fermentation, Yamanashi University, Kitashin 1-13-1, Kofu, 400 Japan. Communicated by Shoji Goto.

The following are abstracts of recently published papers.

Classification of Fragrant Odor Producing Cladosporium species. S. Goto, H. Yamakawa and I. Yokotsuka. J. Agri. Chem. Soc. Japan, 49, 377-381 (1975).

Classification of forty-one fragrant odor-producing Cladosporium species, isolated from various fruit and vegetable trashes, was studied. All isolates produced a marked fruity odor (like ripe bananas, apples, etc.). Four groups were identified as follows: (a) five strains of Cl. cladosporiodes (white colony type), (b) nineteen of Cl. cucumerinum (white colony type), (c) nine of Cl. resinae f. albidum (white colony type) and eight of Cl. fermentans sp. nov. Cl. fermentans has remarkable characteristics in that it forms white colonies, forms clusters of short branched small conidia and it ferments glucose, galactose, sucrose and maltose well.

Classification of Fragrant Odor-Producing Geotrichum. S. Goto, H. Yamakawa and I. Yokotsuka. J. Agric. Chem. Soc. Japan, 49, 519-525 (1975).

Classification of sixty five strains isolated as fragrant odor-producing Geotrichum strains from various fruit and vegetable trashes, was studied. All isolates produced a marked fruity odor. Morphological, cultural and physiological properties of the isolates were examined. The isolates were divided into four groups by morphological characters. These four groups also were distinguished by some physiological characters on fermentation, assimilation of carbon compounds and vitamin requirements. Consequently, some physiological characters in addition to morphological characters were used as criteria for their classification and the isolates were identified as follows: 51 strains (A and B groups) of G. candidum, one (C group) of G. fici sp. nov.. and 12 (D group) of G. rectangulatum sp. nov.. The 51 strains identified as G. candidum were divided into 11 vitamin-requiring strains (B group) and 40 vitamin-independent strains (A group) by the pyridoxine requirement test. G. fici is characterized by long cylindrical arthrospores, formation of irregular short terminal or lateral branches of the hyphae and distinctive fermentation and assimilation of carbon compounds. G. rectangulatum was characterized by big rectangular arthrospores, formation of chlamydospores and distinctive fermentaion and assimilation of carbon compounds.

n-Paraffin Assimilating Yeasts Grown at 37°C. K. Ueno, Y. Asai, M. Shimada and S. Goto. J. Ferment. Technol., 52 861 (1974).

Among 284 n-paraffin assimilating yeast strains isolated from soil, 21 strains showed good growth in n-paraffin medium at comparatively high temperature (37 C) in shaking culture. One of them gave 95.5% cell yield

against n-paraffin added to the medium and the cells had a protein content of 61.8% in jar fermenter studies. Taxonomical studies showed that all of the 21 strains belonged to the genus Candida, including one new species. These are 1 strain of C. kofuensis nov. sp., 12 of C. tropicalis, 2 of C. albicans, 2 of C. solani, 1 of C. krusei, 1 of C. intermedia, 1 of C. rugosa and 1 of C. parapsilosis.

A strongly ethanol assimilating new yeast, Candida brassicae nov. sp. Y. Amano, S. Goto and M. Kagami. J. Ferment. Technol. 53, 311 (1975).

A new yeast, which strongly assimilates ethanol was isolated from cabbage trash. The new yeast, Candida brassicae, is described on the basis of commonly employed taxonomic characteristics. The yeast fermented only glucose and assimilated the six main sugars (glucose, galactose, sucrose, maltose, raffinose and xylose), two alcohols (ethanol, and glycerol) and two organic acids (DL-lactic acid and succinic acid). Potassium nitrate is not assimilated. The new yeast grew at 43-45°C and assimilated ethanol well at the highest temperature.

XXVI. Technical Research Centre of Finland, Biotechnical Laboratory, Bulevardi 29-31, P. O. Box 192, SF-00121 Helsinki 12, Finland. Communicated by T-M. Enari.

Acetate, propionate, sorbate, sugar and fat tolerance of some baker's yeast (Saccharomyces cerevisiae Hansen) strains and of a yeast strain isolated from sour dough (Torulopsis holmii Lodder). by M-L. Suihko and V. Makinen. Technical Research Centre of Finland, Biotechnical Laboratory, Report N:o 11 (pp. 37, in Finnish)

In acetate concentrations of 0.05%, 0.10%, 0.20% and 0.50% the relative fermentation rates of fermentation by 17 different baker's yeast strains fell to average values of 96%, 94%, 91% and 56%. Using a sour dough yeast the respective figures were 102%, 99%, 99% and 79%. In dough of normal pH (5.9) acetate did not inhibit fermentation. When the pH was reduced with lactic acid to 4.3 acetate strongly inhibited fermentation by baker's yeast but not by sour dough yeast. The relative raising power of the baker's yeast strains fell to 33%, while that of the sour dough yeast was the same as in the control experiment. The reduction of pH did not inhibit the raising power.

With propionate concentration of 0.02%, 0.05%, 0.10% and 0.30%, the average relative fermentation rate by the baker's yeast strains fell to 91%, 76%, 64% and 44% respectively. For the sour dough yeast corresponding figures were 101%, 101%, 96% and 71%. In raising tests propionate was added to the extent of 0.16% of the weight of flour. The relative raising power of the baker's yeast strains fell to 88%, while that of sour dough yeast was measured as 105%.

Sorbate concentrations of 0.01%, 0.02%, 0.05% and 0.10% were used. Relative fermentation rates for the baker's yeast strains were on average 88%, 76%, 58% and 42%, while for sour dough yeast they were 95%, 93%, 75% and 60%. In raising tests sorbate was used at a level of 0.08% of the weight of flour. The relative raising powers for the baker's yeast strains and sour dough yeast were 68% and 81%, respectively.

In testing the effect of sugar on the raising power of the yeasts, sugar was added at 14.3% of the weight of flour. The relative raising power of the baker's yeast strains fell to 26-84% and of sour dough yeast to 71%.

Using 25% by weight of fat and sugar, the relative raising power of the baker's yeast strains fell to 12-97% and of sour dough yeast to 66%.

XXVII. Bass Charrington Ltd. High Street, Burton-on-Trent, England. Communicated by B. Ferguson.

The following is a summary of a paper read at the Annual Convention of the American Society of Brewing Chemists, May 1975.

Rapid Detection of Yeast cells using a Radio tracer Technique.

T. C. Box and B. Ferguson

A radiotracer method for the estimation of small numbers of yeast cells is described. The method is based upon the production of $^{14}CO_2$ from D(U-^{14}C glucose) by metabolizing cells.

Yeast cells, either in suspension or isolated on a membrane, are incubated at 26°C in a rich nutrient medium (2 ml) containing radioactive glucose. At the end of fermentation the incubation vial is connected to an absorption train. Sterile air is drawn through the system, so that CO_2 produced during the fermentation is washed from the incubation vial and is quantitatively trapped in an absorbent scintillator solution contained in a scintillation vial. Several samples may be run simultaneously from the same vacuum manifold. The activity of the $^{14}CO_2$ is measured in a liquid scintillation counter with an efficiency of 30 $\pm$ 3%. A counting rate equal to twice that of the background is taken as positive indication of yeast activity in the incubation mixture. A procedure is described for reducing the background count due to non-metabolically produced $^{14}CO_2$: this markedly increases the sensitivity of the method.

The method will readily detect some 25 cells in eight hours and less than five cells in 24 hours. A linear relationship is shown between level of contamination and $^{14}CO_2$ activity.

XXVIII. Institute of Microbiology of the Bulgarian Academy of Sciences, Sofia, Bulgaria. Communicated by V. Kostov.

Physiologic and Biochemical Studies on the Pigment-forming yeasts from the Genus Rhodotorula. III. An Investigation into the Production capacity of a Rhodotorula glutinis strain. V. Kostov, V. Abadjieff, R. Pavlova, L. Krastev, A. Atev, R. Rebarkovska.

Yeasts belonging to the genus Rhoodotorula have been studied in different aspects. The research concerning the abilities of these species of yeasts to accumulate biomass with high percentages of protein and carotenoids, suitable for the needs of animal breeding, are still insufficient. Considering this, the aim of our investigations was to study different nutrient media and to determine the percentage of protein and carotenoids in the biomass.

The experimental strain Rh. glutinis 197 was studied by cultivation in molasses nutrient medium and in a medium of wood hydrolysate in laboratory fermentors of 10 l at 30 C with aeration of 1 liter/min. The total cell count was followed microscopically, the percentage of the budding cells, the viable cells by dehydrogenation of methylene blue after Fink, contamination, reducing sugars after Berthran, absolute dry yeasts (ADY) by weight and percentage of protein after Kjeldal-Nessler. The percentage of carotenoids of certain samples was checked spectrophotometrically and the percentage of amino acids of the same with an amino acid analyser.

The results show that the duration of cultivation in fermentors with equal ratio of the quantity of nutrient medium and inoculating materials is approximately one and the same. With a higher percentage of inoculating yeasts (50%), taken as to the percentage of sugar in the medium, the fermentation takes 24 hours, and at a lower one (30%)-36 hours. By cultivation at 30 C at pH 4.0, aerating and maintaining high sterility, the percentage of live cells varies from about 92-100%. The yield of ADY with respect to the added sugar in the experiment with small quantities of nutrient medium (3,0-4 liter) and a short period of cultivation (-24 - 36 h) oscillated between 18.58 - 42.05. The total quantity of protein was 35.01 - 41.67%. By almost continuous cultivation for 65 hours, the culture develops intensively and the yield of ADY is 48.5 - 49.3% based on the quantity of added sugar. The protein content was 44.88 - 48.13%. The percentage of water in the biomass of yeasts after centrifuging is 72-80%. When the development is active about 40% of the cells bud. The level of carotenoids reaches up to 288 µg/g of ADY.

On hydrolysate medium the fermentation process is longer. Although cell multiplication is considerable, a great part of them (-5-24%) lose viability, probably due to harmful admixtures in the medium - furfurol, etc. There are about 25% budding cells. The yield of ADY based on the added sugar varies (from 18.0 to 42.60%). The quantity of carotenoids reaches 127 µg/g ADY. The percentage of essential amino acids in the biomass is normal - lysine - 3.65% methionine - 0.74% and Tryptophan-0.48%. There is a high percentage of histidine - 3.47%.

On a hydrolysate medium, the quantity of lysine was 3.88%, methionine 0.74% and Tryptophan - 0.48%.

Other authors have reported lower results, biomass 17.4-28.8% containing 23.8 to 29% protein.

From the present studies we concude that on finding a suitable strain and nutrient medium (molasses) and with optimal cultivations, there can be obtained for this kind of yeast maximum yield of biomass with a high content of protein and essential amino acids. Another valuable property of the pigment-producing yeasts is the contents of carotenoids, which are provitamins of vitamin A.

XXIX. Department of Biology. National University Chonnam, South Korea 500. Communicated by M. S. Park.

Studies on the Production of Fermented Feed with Wild Yeasts and Animal Breeding. Communicated by Myung Sam Park.

Abstract

A collection of 96 yeasts was made from various habitats. The best five of them were selected for the production of fermented feed, and also for the industrial utilization of yeast itself and its enzymes. The substrates for fermented feed production were defatted rice, barley, wheat bran, and also sawdust.

A mixture combined the substrates above into varied rations (A, B, C, D, and E) was incoluated with the selected yeasts. The growth rate of the animal was assayed through breeding. The results were as follows.

1. The best 5 strains of yeast selected for feed production were Hansenula anomala var. anomala (No. 225), Candida utilis (No. 400), Candida pelliculosa (No. 405), Debaryomyces hansenii (No. 416-C) and Irpex lacteus (No. 403-A).

2. The optimum pH, and sugar concentration of the yeasts in liquid culture were 5.0 to 5.5 and 10° Balling. The optimum temperature was 35 C for No. 403-A and 225; 30 C for 400, 405 and 416-C. The No. 225 and 403-A grow at a higher temperature than 35 and 40 C.

3. The No. 225 and 403-A had large vegetative cells. The No. 225, 400 and 416-C assimilated cellobiose, xylose and soluble starch. Only No. 416-assimilated inulin. The No. 225, 400 and 403-A assimilated KNO_3 and KNO_2.

4. The No. 403-A strains were yeast-like fungi and showed cellulase activity which might help the propagation of other yeasts on the brans.

5. The combination ratio of bran was as follows: A) mixed bran only, B) defatted rice bran and barley bran (1:1), C) defatted rice bran and what bran (1:1), D) defatted rice bran, what bran and barley bran (1:1:1) and E) defatted rice bran, wheat bran and sawdust (1:1:1).

6. The fermented feeds were prepared by mixing brans, 0.3% ammonium sulfate and 5% (w/w) inoculation of yeast suspension in 4% glucose solution. Water content 60-70%, fermentation temperature 25 - 30 C, and fermentation period 2 to 3 days. The mixed bran combined raw materials into varied ratio improved the quality of fermented feed.

7. The increase in protein content of fermented mixed bran showed the following order: A) 4.46% in only mixed bran, B) 2.81% in defatted rice-barley bran (1:1). C) 6.12% in defatted rice-wheat bran, D) 2.51% in defatted-wheat-barley bran (1:1:1). and 2.55% in defatted rice-wheat-sawdust.

8. The animal breeding experiment, using 180 heads of male Starcros, was made for 8 weeks to compare the value of nonfermented with fermented feed.

9. Total weight of animal per day showed an increase of 10.47g from 35.67g at initiation to 587.03g after 8 weeks.

10. The quantity of animal feed intake was greater in the fermented feed, than in the nonfermented feed.

11. The fermented feed prepared in this experiment by the author might work as a nutritive feed using microbial cell body, enzymes produced by microbes and other microbial cell constituents respectively.

XXX. National and International Meetings on Yeast.

1. The 5th Specialized Symposium on Yeasts will deal with Systematics and Related Problems and will be organized by Dr. Novak and colleagues. The Symposium is to be held at the end of August or at the beginning of September 1977. The place will be Keszthely, Hungary, a small university town situated at the Balaton. Further information will be published later.

2. The Second International Mycological Congress will be held at the University of South Florida, Tampa, Florida, U.S.A. from Saturday, August 27 to Saturday, September 3, 1977. Efforts will be made to organize one or more symposia on yeasts. To obtain a copy of the First Circular interested persons should write to the Secretary of the Congress: Dr. Melvin S. Fuller, Department of Botany, University of Georgia, Athens, Ga. 30602, U.S.A.

3. The Eighth Annual Meeting of the Yeast Genetics Conference-Japan.

The Eighth Annual Meeting of the Yeast Genetics Conference-Japan was held on October 1 and 2, 1975 at the Department of Biology, Nara Women's University, Nara, Japan. Around eighy yeast researchers met, and the following topics were presented and discussed.

Session 1. Mutation and Radiation Effects. Chairmen: T. Ito and I. Takano.

1. T. Takahashi (Suita Lab., Brew. Sci. Res. Inst., Deguchi-cho 5-3, Suita 564) - Genetic effects of p-fluorophenylalanine on Saccharomyces cerevisiae.

2. K. Hieda (Biophys. Lab., Rikkyo Univ., Nishiikebukuro, Toshima-ku, Tokyo 171) - Genetic change induced by partial dehydration in yeast.

3. K. Kobayashi and T. Ito Inst. Phys., Col. Gen. Educ., Univ. Tokyo, Komaba, Meguro-ku, Tokyo 153) - Cell killings and induction of gene conversion by hydroxyurea treatment at different cell stages.

4. T. Ito and K. Kobayashi (Inst. Phys., Col. Gen. Educ., Univ. Tokyo) - The role of singlet excited oxygen molecules in the photodynamic action on yeast cells.

5. T. Saeki, I. Machida and S. Nakai (Div. Genet., Natl. Inst. Radiol. Sci., Anakawa-cho, Chiba 280) - Genetic characters of X-ray sensitive (rad) mutants in yeast.

Session 2. Cytoplasmic Inheritance and Drug Resistance. Chairmen: H. Mori and H. Tamaki.

6. Kota Suda, M. Hakozaki and A. Uchida* (Biol. Lab., Nara Univ. Educ., Nara 620 and* Biol. Div., Col. Gen Educ., Kobe Univ.) - Effects of caffeine on cytoplasmic inheritance in S. cerevisiae.

7. N. Gunge (Central Res. Lab., Mitsubishi Chem. Ind. Ltd., 1000 Kamoshida, Midori-ku, Yokohama 227) - The effect of gene dosage on the transmission and recombination of mitochondrial drug resistance markers.

8. Kayoko Suda and S. Nagai (Dept. Biol., Fac. Sci., Nara Women's Univ., Nara 630) - Genetics of Blasticidin S resistance in S. cerevisiae.

9. T. Yamazaki and Y. Ohara (Dept. Ferment. Technol., Yamanashi Univ., Takeda, Kofu 400) - Induction of drug resistant mutants in Saccharomycodes ludwigii.

10. H. Mori (Noda Inst. Sci. Res., 399 Noda, Noda 278) - Hybridization between salt-tolerant and salt-sensitive strains of Saccharomyces rouxii.

11. H. Tohoyama and T. Murayama (Biol. Inst., Ehime Univ., 2-5 Bunkyo-cho, Matsuyama 790) - Genetical study of cadmium resistant yeast.

12. K. Wakabayashi (Fac. Med., Univ. Tokyo, Hongo 1-7-3, Bunkyo-ku, Tokyo 113) - The localization of oligomycin resistance in a fragment of mitochondrial genome.

13. N. Kane and S. Nagai (Dept. Biol., Fac. Sci., Nara Women's Univ., Nara 630) - Respiratory-deficient mutants in Saccharomycodes ludwigii.

Session 3. Gene Regulation. Chairman: T. Takahashi.

14. H. Tamaki (Doshisha Women's Col., Imadegawa Teramachi, Kamikyo-ku, Kyoto, 602) - Genic analysis of the genes for the fermentation of starch in Sacchaormyces.

15. Y. Nogi, K. Matsumoto and Y. Oshima (Dept. Ferment. Technol., Osaka Univ., Yamadakami, Suita 565) - A new class of gali^{S} mutation producing repressor that represses c mutation in Saccharomyces cerevisiae.

16. K. Matsumoto, Y. Nogi and Y. Oshima (Dept. Ferment. Technol., Osaka Univ.) - Characterization of the temperature sensitive gal4 mutants in Saccharomyces cerevisiae.

Session 4. Cytology and Function of Cell Organelles. Chairman: A. Yuasa.

17. T. Ito and K. Kobayashi (Inst. Phys. Col. Gen. Educ., Univ. Tokyo, Komaba, Meguro-ku, Tokyo 153) - A photodynamic dye probe into

the physiological state of the cell: *In vivo* evidence of phase transition in the membrane structure.

18. M. Osumi (Dept. Biol., Japan Women's Univ., Mejirodai, Bunkyo-ku, Tokyo 112) - Nucleic acid in yeast microbody.

19. K. Takeo (Dept. Bacteriol., Chest Disease Res. Inst., Kyoto Univ., Sakyo-ku, Kyoto 606) - Comparison of cell wall ultrastructure of yeasts by mean of freeze-etching.

20. F. Honda, N. Kane, S. Nagai and K. Takeo* (Dept. Biol. Nara Women's Univ., Nara 630* and Dept. Bacteriol., Chest Disease Res. Inst., Kyoto Univ.) - Effect of Blasticidin S on cellular fine structures of *Saccharomyces cerevisiae*, *Saccharomyodes ludwigii* and *Candida albicans*.

Session 5. Regulation of Vegetative and Reproductive Growth. Chairmen: N. Yanagishima and Y. Oshima.

21. I. Takano (Ctr. Res. Inst., Suntory Ltd., 475 Hirose, Shimamoto-cho, Mishima-gun, Osaka 618) - Genetic analyses of a sporogenous diploid homozygous for α mating-type in *Saccharomyces*.

22. S. Harashima and Y. Oshima (Dept. Ferment. Technol., Osaka Univ., Yamadakami, Suita 565) - Mapping of the homothallic genes in *Saccharomyces* yeasts.

23. Y. Kawanabe, K. Yoshida and N. Yanagishima (Biol. Inst., Fac. Sci., Nagoya Univ., Chikusa-ku, Nagoya 464) - Dependency of sexual aggutinability on cell cycle and culture conditions.

24. M. Hagiya, K. Yoshida and N. Yanagishima (Biol. Inst., Fac. Sci., Nagoya Univ., Chikusa-ku, Nagoya 464) - Purification and molecular complex formation of mating factors which are released from cell walls of *Saccharomyces cerevisiae* by a new mass isolation method.

25. C. Shimoda, Y. Matsushima* and N. Yanagishima** (Dept. Biol., Fac. Sci., Osaka City Univ., Sumiyoshi-ku, Osaka 558,* Dept. Pharmacol., Osaka Col. Pharm. and** Biol. Inst., Fac. Sci., Nagoya Univ.) - Purification and characterization of a cytoplasmic factor inactivating sexual agglutinability in *Saccharomyces cerevisiae*.

26. M. Tsuboi (Dept. Biol., Fac. Sci., Osaka City Univ., Sumiyoshi-ku, Osaka 558) - The role of ribonuclease in sporulation of *Saccharomyces cerevisiae*. Intracellular localization of ribonuclease.

27. S. Doi and M. Hayashibe (Dept. Biol. Fac. Sci., Osaka City Univ., Sumiyoshi-ku Osaka 558) - Effect of temperature shock on bud initiation in *Saccharomyces cerevisiae*.

28. M. Hayashibe and K. Tanaka* (Dept. Biol. Fac. Sci., Osaka City Univ. and* Inst. Appl. Microbiol., Univ. Tokyo) - Changes in cellular fine structure during induction period for budding in *Saccharomyces cerevisiae*.

Session 6. Metabolism and Biochemical Aspects. Chairmen: T. Kamihara and M. Dohi.

29. A. Kimura, M. Okuda, K. Hirose and S. Nagai* (Dept. Food Sci. Technol., Fac. Agr., Kyoto Univ., Kyoto 606, and* Dept. Biol. Fac. Sci., Nara Women's Univ.) - Analysis of control mechanism of ATP-generating system in nucleic acid fermentation by the use of respiration-deficient mutants of yeasts.

30. M. Hashimoto, H. Mori** and A. Kimura (Dept. Food Sci. Technol., Kyoto Univ., Kyoto 606 and* Noda Inst. Sci. Res.) - Control mechanism of ATP-generating system in a temperature-sensitive mutant of a yeast, Saccharomyces rouxii.

31. Kenji Arima, M. Hashimoto and A. Kimura (Dept. Food Sci. Technol., Fac. Agr., Kyoto, Univ.) - The role of outer membrane of yeasts in nucleic acid fermentation.

32. I. Nakamura, Y. Nishikawa, T. Kamihara and S. Fukui (Lab. Indust. Biochem, Dept. Indust. Chem., Fac. Engineer., Kyoto Univ., Kyoto 606) -Respiratory adaptation with pyridoxine in Saccharomyces carlsbergensis grown with thiamine.

33. K. Doi (Inst. Sci. Indust. Res., Osaka Univ., Yamadakami, Suita 565) - Differential behaviour of the components of Arthrobacter β-1, 3 glucanases on Avicel column.

34. K. Doi (Inst. Sci. Indust. Res., Osaka Univ.) - A simple device for raising reproducibility in fluorometric measurement of deoxyribonucleic acid in yeast cells.

35. T. Mizunaga and M. Nakamura (Dept. Agr. Biol. Chem., Univ. Tokyo, Bunkyo-ku, Tokyo 113) - Some properties of constitutive and repressible acid phosphatases of baker's yeast.

36. M. Dohi and Kei Arima (Dept. Agr. Chem., Fac. Agr., Univ. Tokyo, Bunkyo-ku, Tokyo 113) - Effect of releasing of cellular components from the cell envelope on the activities of phosphatases in inositol-deficient yeast cells.

Yeast researchers interested in these papers are welcome to contact the author. The next annual meeting will be held in Tokyo in August, 1976.

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XXXI. Brief News Items

1. Czechoslovak yeast specialists associated in the Czechoslovak Commission for yeasts of the Czech. microbiological society wish to all yeast specialists, friends and their families on the world very happy Christmas, much personal health, success in research and the whole world peace in the year 1976. Anna Kockova-Kratochvilova

2. The 1975 edition of the catalogue of the British National Collection of Yeast Cultures is now available, price £1.00. Barbara Kirsop, Curator, The Brewing Industries Research Foundation, Nutfield, Redhill, Surrey RH1-4HY, England.

3. The following paper has recently been published: The genus Torulaspora Lindner. J. P. van der Walt and E. Johannsen. Microbiology Research Group, Bulletin 2. CSIR Res. Rept. No. 325 pp. 1-23, 1975. Copies of this report may be obtained from J. P. van der Walt, P. O. Box 395, Pretoria 001, South Africa.

4. The following paper has been published recently: Mossbauer Effect and Electron Paramagnetic Resonance Studies on Yeast Aconitase (Candida lipolytica). T. Suzuki, Y. Maeda, H. Sakai, S. Fujimoto and Y. Morita. J. Biochem. 78, 555-560, 1975. Takashi Suzuki, Takeda Chemical Industries, Ltd., Takasago Plant, Takasago, Hoogo 676, Japan.

5. The titles of two recent papers from our laboratory are: A Possible Mechanism of Energy Coupling in Purine Transport of Saccharomyces cerevisiae, Uwe Reichert, Rainer Schmidt and Martine Foret, Volume 52, No. 1, pg. 100-102, FEBS Letters, March 1975.

Uptake and Accumulation of Purine Bases by Stationary Yeast Cells Pretreated with glucose, Uwe Reichert and Margrit Winter, Biochimica et Biophysica Acta, 356(1974) 108-116. Uwe Reichert, Freir Universitat Berlin, Zentralinstitut fur Biochemie und Biophysik, Haus V, 1 Berlin 33, Ehrenbergstrasse 26-28.

6. The following papers have recently been published from our laboratory:

Fermentative Production of CDP-Choline and Related Cytidine Coenzymes by Dried Cells of Yeasts, Akira Kimura, Yashuiro Kariya, Kazuo Aisaka, and Tatsurokuro Tochikura, Proceedings of the first intersectional congress of IAMS, Vol. 5, 510 (1975).

Fermentative Production of CDP-Choline Analogues by Hansenula jadinii, Yasuhiro Kariya, Kazuo Aisaka, Yoshinobu Kaji, Akira Kimura and Tatsurokuro Tochikura, Reprinted from Journal of Fermentation Technology, Vol. 53, No. 8 (1975).

Fermentative Formation of CDP-Choline by Intact Cells of a Yeast, Saccharomyces carlsbergensis (IFO 0641) Treated with a Detergent, Triton X-100, Akira Kimura and Makoto Morita, Agr. Biol. Chem., 39(7) 1469-1474, 1975.