

Y E A S T

A News Letter for Persons Interested in Yeast

January 1969

Volume XVII, Number 2

Editor

Herman J. Phaff, University of California, Davis, California

Associate Editor

Leslie R. Hedrick, Illinois Institute of Technology, Chicago, Illinois

Associate Editor

F. M. Clark, University of Illinois, Urbana, Illinois

Associate Editor

Cecil G. Dunn, Massachusetts Institute of Technology, Cambridge, Massachusetts

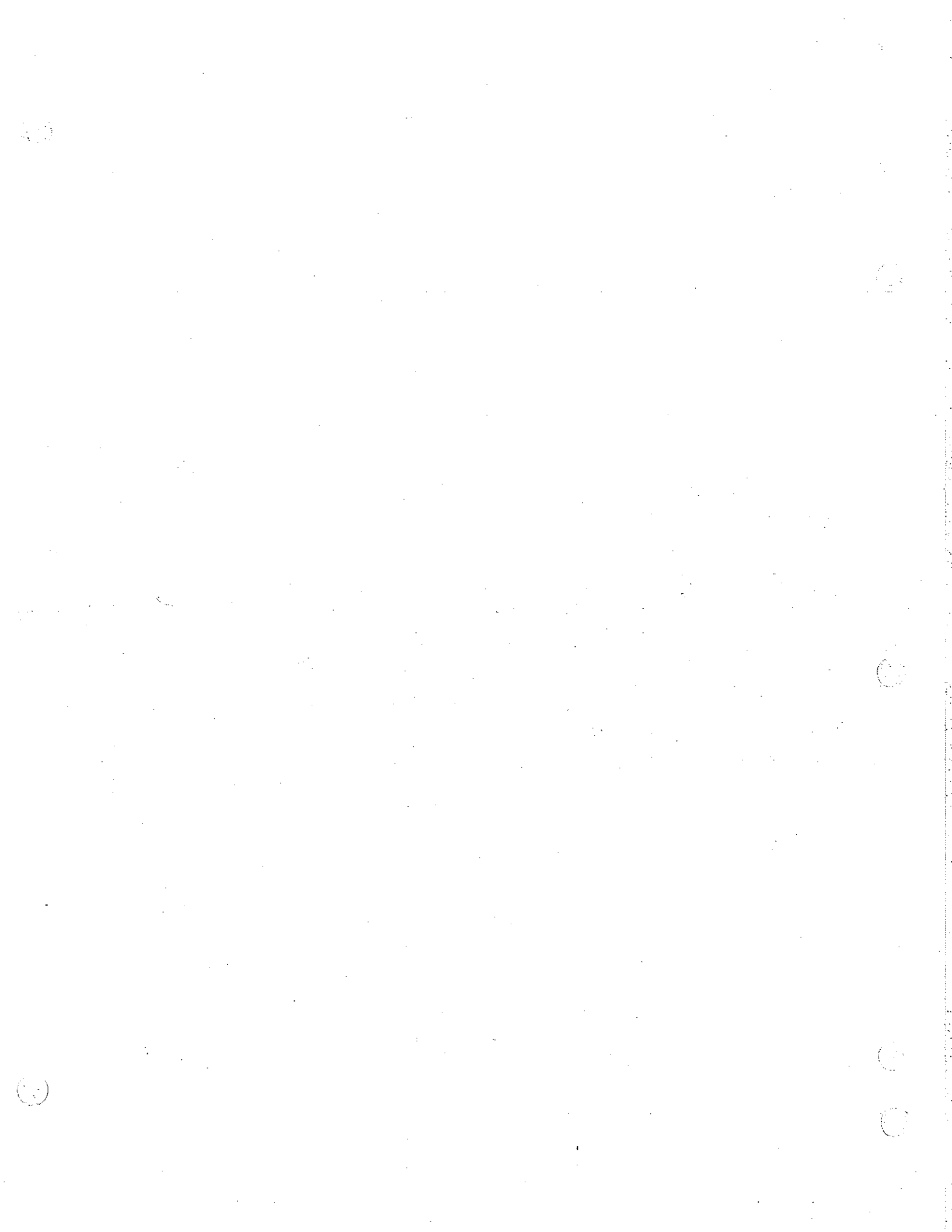
	<u>Page</u>
D. Yarrow, Delft, The Netherlands	27
Shoji Goto, Kofu, Japan	27
T. Nakase, Kawasaki, Japan	28
D. G. Ahearn, Atlanta, Georgia	29
Leo Kaufman, Atlanta, Georgia	29
E. Minárik, Bratislava, Czechoslovakia	30
O. Verona, Pisa, Italy	30
M. Kozaki, Tokyo, Japan	31
Edward J. Buecher, Jr., Berkeley, California	32
S. P. Meyers, Baton Rouge, Louisiana	32
Richard Snow, Davis, California	33
J. R. Johnston, Glasgow, Scotland	34
N. Yanagishima, Osaka, Japan	35
A. Sols, Madrid, Spain	36
Heikki Suomalainen, Helsinki, Finland	37
Pamela A. D. Rickard, Kensington, Australia	40
B. C. Rankine, Adelaide, South Australia	42
A. H. Cook, Surrey, England	42
S. Windisch, Berlin, Germany	43
International Meetings	43
Brief News Items	45

Many thanks to those who have contributed to this issue by sending in news items and accounts of research projects. The next issue will be published in May 1969. A contribution of \$1.00 from those who have not contributed for some time would be appreciated to finance future editions of the News Letter. Many thanks to those who have contributed recently.

The Editor regrets that due to a number of unforeseen circumstances this issue will arrive late; in particular so because of a surface mail embargo to European and African countries due to a dock workers strike in the eastern United States.

The Editor extends to the readers of the Yeast News Letter his warmest wishes for a happy and productive new year ahead.

H. J. Phaff



I Centraalbureau voor Schimmelcultures, Delft, Julianalaan 67a. Communicated by Mr. D. Yarrow.

The following new species have been received in the collection since the appearance of the last issue of the Yeast News Letter.

Debaryomyces halotolerans CBS 5949

Y. Sasaki & T. Yoshida, Jap. J. Ferm. Technol. 44: 61-71 (1966).

Debaryomyces nepalensis CBS 5921

S. Goto & J. Sugiyama, J. Jap. Bot. 43: 102-110 (1968).

Pichia krusei CBS 5945 (= IFO 0035)

T. Tsuchiya, Y. Fukuzawa, T. Shinoda & M. Imai, Jap. J. Exp. Med. 37: 285-290 (1967).

Rhodotorula vuilleminii CBS 5951

H. Saéz, Bull. Soc. Mycol. France 83: 953-958 (1968).

Saccharomyces hispanica CBS 5835

J. Santa María, Bol. Inst. Nac. Invest. Agronóm. 58: 21-32 (1968).

A revised, 1968, edition of the list of cultures held by the CBS is now available. This can be obtained from the Centraalbureau voor Schimmelcultures, Oosterstraat 1, BAARN, the Netherlands and the price is D.Fl 10.- per copy.

II Institute of Fermentation, Yamanashi University, Kitashimachi, Kofu, Japan. Communicated by Dr. Shoji Goto.

The classification of yeasts isolated from various samples at the uplands in Himalayan districts has been studied during the last few years.

1) Coprophilous fungi from the Karakorum.

J. Sugiyama and S. Goto: J. Japanese Botany, 42: 75 (1967)

Seven species of coprophilous fungi, including one new species, i.e., Sporobolomyces coprophilus and one new variety, i.e., Cryptococcus albidus var. ovalis, were reported from goat dung collected in Hisper Valley, Karakorum, Pakistan.

Aspergillus candidus and Crypt. albidus show a world-wide distribution on animal dungs.

The following fungi, i.e., Stemphylium ilicis, Sp. coprophilus, Crypt. neoformans, Crypt. albidus var. ovalis and Rhodotorula marina, were recorded for the first time as inhabitants of the dung mycoflora.

Sp. coprophilus hereby described differs from Sp. roseus in having spores which are oval to long oval and in galactose not being assimilated.

2) Studies on Himalayan yeasts and moulds.

S. Goto and J. Sugiyama: J. Japanese Botany, 43: 102 (1968)

I. A new species of Debaryomyces and some asporogenous yeasts.

Six yeasts were isolated from various samples at East Nepal. They are identified as follows: 2 strains of D. nepalensis n. sp., 2 of Candida melinii, 1 of Rhodotorula marina and 1 of Rh. rubra.

D. nepalensis can definitely ferment glucose, sucrose and raffinose, but not galactose, maltose and lactose. These isolates can not assimilate lactose and require biotin for growth. C. melinii - Himalayan strain - has the characteristic that it can grow between 15-34 C. In Rh. rubra - Himalayan strain, the authors discovered that

unusual cells, namely septate cells with one septum, appeared in a colony of six months old.

3) Studies on Himalayan yeasts and molds.

S. Goto and J. Sugiyama: in press.

II. New species of Cryptococcus and Candida and some asporogenous yeasts.

Thirteen yeasts, six species including three new species Crypt. superioa, Crypt. himalayaensis and Candida montinus, were isolated from soils, dung and humus that were collected in Himalaya. They are all nonfermenting asporogenous yeasts; 2 strains of Crypt. diffluens, 1 of Crypt. superioa, 2 of Crypt. himalayaensis, 4 of Torulopsis candida, 2 of C. montinus and 2 of Rhodotorula glutinis.

Crypt. superioa is characterized by its inability to assimilate inositol, and Crypt. himalayaensis is characterized by inability to assimilate maltose and ability to assimilate potassium nitrate and lactose. Candida montinus is characterized by ability to form septate mycelial cells, to produce starch-like substances and to assimilate melibiose.

III Ajinomoto Co., Inc., Central Research Laboratories, 2964, Suzuki-cho, Kawasaki, Japan. Communicated by Dr. T. Nakase.

1. The following paper will be published in the December issue of J. Gen. Appl. Microbiol. (14, 345-357, 1968):

"Taxonomic significance of base composition of yeast DNA" by T. Nakase and K. Komagata

2. The following article has been accepted for publication in J. Gen. Appl. Microbiol.:

"DNA base composition of the genus Hansenula" by T. Nakase and K. Komagata

Summary: The DNA base ratio of 46 cultures of Hansenula and allied yeasts which represent 26 species or varieties was studied. The GC content of Hansenula DNA ranged from 28.5 to 46.3%. Intrageneric variation ranged from 15.6-17.8% in Hansenula sensu Wickerham, 14.4-15.8% in Hansenula sensu Lodder and Kreger-van Rij, 13.4-14.8% in Hansenula emended by Novak and Zsolt, and 11.0-12.7% in Hansenula group (genus) by Tsuchiya et al. The frequency curve of the DNA base ratios demonstrated the presence of two large groups of the species in the genus Hansenula. The first group had relatively high values of GC contents with maximal frequency in the class of 40-42%. The second had relatively low values of GC contents with the maximal frequency in the class of 32-34%. Species arranged in each line of the phylogenetic scheme of Hansenula proposed by Wickerham exhibited consistent DNA base ratios, respectively, though exceptions were found. So did the serological groups within Hansenula group by Tsuchiya et al. A taxonomic and phylogenetic evaluation of DNA base ratios was discussed in relation to the delimitation of the genus Hansenula by several authors and the existence of several groups having similar DNA base ratios within the genus.

IV Department of Biology, Georgia State College, Atlanta, Georgia, 30303.
Communicated by D. G. Ahearn.

The following papers recently have been published:

Ecology and characterization of yeasts from aquatic regions of South Florida. D. G. Ahearn, F. J. Roth and S. P. Meyers. *Marine Biology* 1(14):291-308. 1968.

Shape and structure of the ascospores of Hanseniaspora uvarum. N. J. W. Kreger-van Rij and D. G. Ahearn. *Mycologia* 60(3):604-612. 1968.

Extracellular proteinases of yeasts and yeastlike fungi. D. G. Ahearn, S. P. Meyers and R. A. Nichols. *Appl. Microbiol.* 16(9):1370-1374. 1968.

During the past August, Dr. S. P. Meyers (Louisiana State University), Dr. W. L. Cook (N. Y. State College, Plattsburgh), Miss Gayle Hansen (University of Vermont) and I participated in an investigation of the yeast flora of Lake Champlain. Yeasts were collected from various depths at 27 stations using Niskin biological samplers; deepest samples were collected at 115 meters. The systematics and certain physiological properties of the strains isolated are currently being investigated.

V Department of Health, Education, and Welfare, National Communicable Disease Center, Atlanta, Georgia 30333. Communicated by Dr. Leo Kaufman.

Diagnostic tests for Cryptococcosis

The Fungus Immunology Unit of the Mycology Section, National Communicable Disease Center, has recently completed an evaluation of a number of serological tests used for the diagnosis of cryptococcosis.¹ The study showed that maximal early and accurate serologic diagnosis of cryptococcosis can be accomplished through the concurrent use of three tests: the latex agglutination (LA) test for cryptococcal antigen, the indirect fluorescent antibody (IFA) test, and the tube agglutination (TA) procedure for Cryptococcus neoformans antibodies.

In the evaluation these tests were applied to 141 serum and cerebral spinal fluid (CSF) specimens from 66 patients with culturally proven cryptococcosis and to 42 sera from normal subjects and from patients with other systemic mycotic diseases. On the basis of these results, the LA test is sensitive and completely specific. Fifty-five percent of the sera from proven cryptococcosis cases were positive. With the TA test, 37% of the specimens were positive, and the test was highly specific. With the IFA test, 38% of the specimens were positive, but the test appeared to be the least specific of the three. Cross-reactions were most evident with sera from patients with blastomycosis or histoplasmosis. When the three tests were used concurrently, 87% of the cryptococcosis specimens were positive, and they permitted a presumptive diagnosis of C. neoformans infections in 61 (92%) of the 66 patients whose specimens were examined.

There appears to be no correlation between clinical types of cryptococcosis and reactivity with any particular serologic test. The level of the TA titer and the intensity of staining in the IFA test were not related to the severity of infection. Cryptococcal antibodies were rarely detected in CSF

specimens (1 out of 21); however, sera from patients with central nervous system involvement did contain demonstrable antibodies.

A positive TA or IFA reaction is presumptive evidence of cryptococcosis. However, with the IFA test particularly, a positive reaction could also reflect a cross-reaction or previous infection.

The LA test is diagnostically useful and appears to be most valuable in detecting cryptococcal meningitis. Although any LA titer appears diagnostic for cryptococcosis, increasing titers seem to reflect progressive infection, and declining titers indicate response to chemotherapy and progressive recovery.

¹Kaufman, L., and S. Blumer. The Value and Interpretation of Serologic tests for the Diagnosis of Cryptococcosis. Applied Microbiol. Dec. 1968.

VI Research Institute for Viticulture and Enology, Bratislava, Czechoslovakia.
Communicated by Dr. E. Minárik.

The following paper has been prepared for publication: "Ecology of yeasts and yeast-like microorganisms of secondary habitats in the vine-region of Tokay".

The yeast flora occurring in typical Tokay cellars in Czechoslovakia was studied. In dry and sweet Tokay wines alcohol-resistant Sacch. oviformis prevails. Film-yeasts of the genus Candida and Hansenula occur in wines aging in incompletely filled casks. Torulopsis, Rhodotorula and Sporobolomyces sp. are very common on cellar walls and floors. They appear as pink, reddish, brown or yellow islets on cellar walls coated with Cladosporium. They are accompanied by Leuconostoc mesenteroides and other lactic bacteria. Mucous yeasts are a regular component of the microflora of cellar equipment but never occur on primary habitats in the vineyard.

At the Annual Plenary Session of the Office International de la Vigne et du Vin/Paris/ held in Bucarest in September 1968 the O.I.V. Price 1968 in Enology was awarded to E. Minárik for the publication: "Ecology of Natural Wine Yeasts Species in Czechoslovakia"/ed. Slovak Academy of Sciences, Bratislava 1966/.

VII Istituto di Patologia Vegetale e Microbiologia Agraria e Tecnica-Universita di Pisa(Italia). Communicated by Prof. O. Verona.

The papers listed below have been published since our last communication to the News Letter:

1. A. A. Lepidi: On the gastro-enteric yeast microflora of the wild edible dormouse (Glis glis L.).

The microflora of the environment under consideration is abundantly constituted by yeasts (Debaryomyces subglobosus, Hansenula saturnus, Saccharomyces drosophilorum, S. rosei, Kloeckera lodderi, Rhodotorula glutinis and occasionally other species).

Some physiological researches showed that, among the isolated strains, prototrophism with regard to vitamins and simple sugars and organic acids assimilation are very prevalent. (L'Agric. Ital., 46, 39, 1968).

2. A. A. Lepidi e G. Picci. On the yeast microflora of the gastro-enteric cavity of the snail (*Helix aspersa* Müller):

I°-Influence of trophic factors.

II°-Effect of environmental temperature.

Research was carried out on the quantitative and taxonomical aspects of the gastro-enteric yeast flora of snails, which had been fasting for various times (0-3-10-20-30 days) and subsequently fed with sterile food (tests were made 3 and 10 days after feeding).

The results showed a remarkable growth of some yeast species (*Debaryomyces hansenii*, *Rhodotorula mucilaginosa*, *Rhodotorula* sp. I and *Torulopsis aerea*) and a specificity of the yeast-microflora in the gastro-enteric organs.

The second paper is concerned with the effect of temperature as a probable factor of the selectivity within the gastro-enteric yeast microflora of the snail.

The temperature results in a quantitative and qualitative action: i.e. an increase in temperature (10-20-30-35°C) causes a diminution both in the average number of cells representing the yeast species and of average frequency of yeast species in the animals. (L'Agric. Ital. 46, 119, 1968; 46, 173, 1968)

VIII Tokyo University of Agriculture, 1-1 Sakuragaoka 1-chome, Setagayaku, Tokyo.
Communicated by Dr. M. Kozaki.

The following is a summary of two studies carried out in our laboratory.

- (1) Yeasts Isolated from Swelled Canned Apple.

MICHIO KOZAKI, NAOHIRO OHARA and KAKUO KITAHARA

Ordinarily, for sterilizing canned apple juice about 76°C flash-ing is used, but there appear frequently somewhat swelled cans due to gas formed by biological infections. From these cases of spoil-age, yeasts were isolated and identified.

All these isolates are referred *Saccharomyces italicus* CASTELLI. Vegetative cells of this species tolerate wet heating at 65°C for 5 minutes and the ascospores tolerate as high as 75°C for 5 min. These values are distinctly higher than those of other species of *Saccharomyces*.

Ratio of asci to total cells is extraordinarily high (e.g. 50%) on ordinary Na-acetate agar medium. From these experiments, we propose to raise the sterilizing temperature to 80C.

- (2) The yeast flora of "Takuan-zuke".

MICHIO KOZAKI and KINSHI SUMINOE

Takuan-zuke is a salted preserved food in Japan. The salted product is prepared by one of the three following general methods (1) by salting dried radish packed in rice bran and dry salt, (2) by salting semidried radish in about 20% salt brine, then repacking in salted rice bran (Honzuke-takuan) and (3) by salting fresh radish with dry salt (Hayazuke-takuan).

The first method is usually done by the housewife and the latter two methods are normally commercial operations. It is well known that these products are ripened by microorganisms, such as lactic acid bacteria and yeasts. We have examined the changes in yeast population in Takuanzuke during 6 months.

Honzuke-takuan was an excellent habitat for Debaryomyces only and the species of isolated yeasts belonged to D. nicotianae, D. kloeckeri and Torulopsis famata.

On the other hand, the genera of yeast isolated from Hayazuke-takuan were much more varied, including Debaryomyces, Hansenula and Saccharomyces.

IX Clinical Pharmacology Research Institute, 2030 Haste Street, Berkeley, Calif. 94703. Communicated by Dr. Edward J. Buecher, Jr.

Caenorhabditis briggsae and other free-living nematodes require a proteinaceous growth factor in order to reproduce and reach high populations. Up until now, this factor had been derived from chick embryo and liver extracts.

Dr. Edward J. Buecher (formerly of Dr. Phaff's laboratory) and Dr. Eder L. Hansen have found that extracts of yeast also will support the reproduction of C. briggsae and other free-living nematodes in bacteria-free (axenic) culture. The discovery was first made using extracts of Saccharomycopsis guttulata, budding strain JB-1, (cf. Yeast News Letter, June 1968). In order to obtain larger quantities of extract, baker's yeast (Fleishmann's yeast cake) was used. Cells were washed once in buffer and broken in either a colloid mill or a Waring blender in the presence of glass beads.

High populations of nematodes were obtained either by using dialyzed yeast extract as the complete medium, or by adding the extracts to a chemically defined medium. Biological activity of extracts was enhanced in the presence of defined medium if they were first completely dialyzed, ammonium sulfate precipitated, or gently heated. Preparations were stable to lyophilization.

X Department of Food Science and Technology, Louisiana State University and Agricultural and Mechanical College, Baton Rouge, Louisiana 70803. Communicated by Dr. S. P. Meyers.

1) Dr. Samuel P. Meyers, formerly of the Institute of Marine Sciences, University of Miami is now a Professor in the Department of Food Science and Technology, Louisiana State University, Baton Rouge, Louisiana, where he will be responsible for development of studies in marine microbiological food research and sea grant estuarine programs. Dr. Meyers will continue to serve as Editor of the Aquatic Microbiology Newsletter (of the American Society for Microbiology), now distributed world-wide to over 800 aquatic microbiologists.

2) Dr. Meyers and Dr. D. G. Ahearn, Georgia State College, Atlanta, Georgia, were participants during the summer at the Miner Center Community of Scholars, Chazy, New York. Work was conducted with Mr. Warren Cook, New York State University College of Arts and Sciences, Plattsburgh, New York, in investigations of the yeasts and molds of Lake Champlain. Miss Gayle Hansen of the Department of Microbiology of the University of Vermont (completing her M.S. degree on portions of Lake Champlain mycology) also worked with this group. An extensive series of collections were made for the first time over the entire Lake, the results of which are now being processed, and the data analyzed for publication early in 1969.

3) The following paper on yeast proteolysis has been published:

"Extracellular Proteinases of Yeasts and Yeastlike Fungi". Applied Microbiology 16, 1370-1374. 1968 (D. G. Ahearn, S. P. Meyers, and R. A. Nichols).

SUMMARY

Approximately 800 yeasts and other fungi, representing over 70 species, were tested for extracellular caseinolysis. Isolates of a variety of genera, including Aureobasidium, Cephalosporium, Endomycopsis, Kluyveromyces, and numerous sporobolomycetes, demonstrated significant proteolytic activity. Caseinolysis was not necessarily correlated with gelatin liquefaction or with albuminolysis. Numerous fungi showed significant proteolysis at 5 C. The most active organisms were isolates of Candida lipolytica, Aureobasidium pullulans, Candida punicea, and species of Cephalosporium. Taxonomic and ecological implications of proteolytic activity are discussed.

XI Department of Genetics, University of California, Davis, Calif. 95616.
Communicated by Dr. Richard Snow.

Work on yeast genetics at this laboratory is currently concerned with three main projects: 1) study of the regulation of acid phosphatase; 2) study of the fine-structure and complementation patterns at the histidine-1 locus; 3) study of radiation-sensitive mutants.

Mr. Jon Kuhn is working with acid phosphatase mutants. He has found that this enzyme is highly inducible, in contrast to many other enzymes in yeast. Derepressed levels are some 200-fold higher than repressed levels. He has isolated several mutants which cannot use β -glycerophosphate as a phosphate source and which lack enzyme activity by both in vitro and in vivo tests. By studying the activity of these mutants toward a number of possible substrates, he has concluded that there is probably only one phosphomono-esterase in the pH 3.0-5.0 range. He is also attempting to isolate constitutive mutants to further the genetic and regulatory studies in progress.

Mr. Christopher Korch has constructed a fine-structure genetic map with about 70 alleles at the locus which controls the structure of the first enzyme in histidine biosynthesis, histidine-1. The fine-structure map was constructed by the X-ray mapping method and also by another method based on the use of methylmethane-sulfonate. The maps constructed by these two methods are generally consistent. In addition he has studied the complementation patterns of the majority of the diploid pairwise crosses of the mutants, and has found that the map must be represented in circular form. He hopes to purify the enzyme sufficiently to obtain electron microscope pictures of it, which may give information on the number of subunits in the active molecule and their relationship to the complementation pattern.

Miss Beverley Reno has been studying mutants which she isolated that are sensitive to X-rays and to methylmethanesulfonate, a chemical whose effects in many ways resemble those of X-rays. She has tested their survival after treatment with X-rays, ultraviolet light, MMS, and nitrous acid, and has found differential responses. She plans to test the effect of these mutants on spontaneous mutation rate, mitotic and meiotic recombination,

and on gene conversion. In addition she is studying the effect of liquid holding and photoreactivation on these mutants and on a number of others which I had obtained previously. Ultimately she hopes to form a hypothesis concerning the spectrum of the repair mechanisms in yeast and to correlate these with their effects on recombination and gene conversion.

My own work is primarily involved with the study of radiation-sensitive mutants. A number of mutants have been isolated which are abnormally sensitive to ultraviolet light, X-rays, nitrous acid, or methylmethanesulfonate. At the moment, 50 such mutants have been found. Current efforts are directed toward mapping the genetic position of the mutants on the yeast chromosomes, to study of the response of some of the double mutants to the above mutagenic agents, and to study of the effects of the mutants on recombination and mutation. An interesting class of mutants has recently been isolated which are sensitive to X-rays and UV at 30 C and which cannot grow at 37 C. These may represent mutation of an enzyme of essential function in normal DNA metabolism which has impaired function at 30 C on irradiated DNA and whose function is completely abolished by high temperatures.

I am also doing some genetic work on the "Montrachet" strain of yeast, a strain which is widely used in winemaking. This strain is diploid and homothallic, single-spore isolates growing into cultures which will sporulate without mating. The process of diploidization of single-spore cultures has been observed microscopally, and occurs soon after the spore germinates. I am isolating a number of auxotrophic mutants in the strain and plan to use these in a genetic analysis of the homothallic response. Some of these auxotrophic mutants, especially those involved in the isoleucine-valine and leucine pathways, should be of interest in the microbiology of winemaking, since the flavor components due to higher alcohol production result from these synthetic pathways. Mr. Frank Bayliss, a graduate student in the Department of Viticulture and Enology on this campus, is presently studying this aspect.

XII University of Strathclyde, Department of Applied Microbiology, Royal College, George Street, Glasgow C-1, Scotland. Communicated by Dr. J. R. Johnston.

At a symposium of the Botanical Society of Edinburgh held in Glasgow in September, Mr. P. V. Patel read a paper entitled "Genetics of nystatin resistance in yeast". (P. V. Patel and J. R. Johnston).

The following is an abstract which will appear in the Transactions of the Society:

"The kinetics of survival of various pathogenic strains of Candida albicans and laboratory-bred strains of Saccharomyces cerevisiae upon exposure to nystatin have been studied. Comparison of killing curves reveals (a) a positive relationship between nystatin-resistance and the ploidy of Saccharomyces strains, (b) the relative resistance of Candida albicans compared to Saccharomyces cerevisiae. Genetic analysis of stable, nystatin-resistant isolates of haploid Saccharomyces strains shows that resistance is due to single gene mutations. Most mutations are dominant (Patel and Johnston, Appl. Microbiol. 16, 164, 1968), but some are recessive as are the mutants of Ahmed and Woods (Genet. Res. 9, 179, 1967). Hybrids of mutants and sensitive strains and segregants of these crosses display various levels of resistance best explained by postulating the presence of modifier genes

(increasing the level of resistance) and suppressor genes (decreasing the level of resistance) in certain of the parent strains. Tetrad analysis suggests that one modifier gene is linked to the basic (resistance) gene. Nystatin mutants were tested for their resistance to "Fungizone" (amphotericin B). Mutants fall into a few distinct groups according to their degree of cross-resistance. This classification assists in identifying the loci to which mutations belong."

XIII Laboratory of Cell Biology, Faculty of Science, Osaka City University, Sumiyoshi-ku, Osaka, Japan. Communicated by Professor N. Yanagishima.

We have been studying hormonal control of yeast growth, using Saccharomyces cerevisiae. The following is a brief summary of research in progress.

A plant hormone, auxin which is produced by the yeast, has been known to be involved in the control of yeast growth.

High concentrations (200-600 mg/l) of auxin (indole-3-acetic acid, α -naphthaleneacetic acid, 2,4-dichlorophenoxyacetic acid) induced in high frequency heritable changes in the cytochrome system, cell form and size and the ability to respond to the cell-expanding action of auxin. The cell-expanding action of auxin on an auxin-responsive variant yeast has important features in common with that on higher plants. Both the action on yeast cells and that on higher plants require special RNA functional in auxin action and are inhibited by antiauxin and inhibitors of RNA and protein metabolism. Auxin is known to control cell differentiation and the ability of cells to respond to the cell-expanding action of auxin differs depending on the degree of cell differentiation in higher plants. Hence the relation between auxin-induced variation in yeast and cell differentiation in higher plants is of interest. An auxin-responsive variant showed irregular segregation in mating ability and changes in chemical nature of the cell wall.

We have succeeded in isolating hormone-like substances from haploid cells of heterothallic strains. These substances, one is excreted by α type cells and the other by α type cells, caused expansion of cells of opposite mating type. Heterothallic diploid cells did not respond to these substances, but diploid cells containing some homothallism-controlling genes responded. These substances seem to be steroidal compounds.

Both auxin and the hormone-like substances are thought to cause cell expansion through activation of wall-degrading enzymes.

The following papers appeared from this laboratory during 1967 and 1968.

1. S. Kamisaka, N. Yanagishima & Y. Masuda. Effect of auxin and gibberellin on sporulation in yeast. *Physiol. Plant.* 20:90-97. 1967.
2. S. Kamisaka, Y. Masuda & N. Yanagishima. Gibberellin-induced yeast sporulation in relation to RNA and protein metabolism. *Physiol. Plant.* 20: 98-105. 1967.
3. S. Kamisaka, Y. Masuda & N. Yanagishima. Yeast sporulation and RNA as affected by gibberellic acid. *Plant & Cell Physiol.* 8:121-127. 1967.

4. N. Yanagishima & C. Shimoda. Production of yeast variants by auxin and effects of plant growth regulators on these variants. *Plant & Cell Physiol.* 8:109-119. 1967.
5. N. Yanagishima. Induction of heritable respiration deficiency in yeast by salt solution. *Plant & Cell Physiol.* 8:211-215. 1967.
6. Y. Masuda, E. Tanimoto, C. Shimoda, S. Kamisaka & N. Yanagishima. Separation of RNA functional in auxin action. *Plant & Cell Physiol.* 8:221-225. 1967.
7. C. Shimoda, Y. Masuda & N. Yanagishima. Nucleic acid metabolism involved in auxin-induced elongation of yeast cells. *Physiol. Plant.* 20:299-305. 1967.
8. N. Yanagishima, C. Shimoda, S. Kamisaka & T. Takahashi. Auxin-induced heritable variants in yeast with special reference to physiological and genetic characters. *Plant & Cell Physiol.* 9:323-331. 1968.
9. N. Yanagishima & C. Shimoda. Auxin-induced expansion growth of cells and protoplasts of yeast. *Physiol. Plant* 21:1122-1128. 1968.
10. C. Shimoda & N. Yanagishima. Strain dependence of the cell expanding effect of β -1,3-glucanase in yeast. *Physiol. Plant.* 21:1163-1169. 1968.

The following paper has been accepted for publication.

11. C. Shimoda & N. Yanagishima. Relation of special RNA to sensitivity of yeast strains to auxin. *Plant & Cell Physiol.* 10: 1969.

XIV Centro de Investigaciones Biologicas, Departamento De Enzimologia, Velazquez, 144 - Madrid - 6 (Spain). Communicated by Dr. A. Sols.

The following two papers have been published recently in the *European Journal of Biochemistry* 5:165 and 321, 1968.

Glycerol Metabolism in Yeasts - Pathways of Utilization and Production.
C. Gancedo, J. M. Gancedo, and A. Sols.

The utilization of glycerol by Candida utilis has been studied. It has been found that this yeast has a permeability for glycerol and other three carbon compounds much greater than that of baker's yeast. This permeability allows the entrance of glycerol in Candida cells rapidly enough to permit its efficient utilization even at low concentrations. The inducibility of glycerol kinase has been established. An increase in the concentration of the mitochondrial L- α -glycerophosphate oxidase when the yeast is grown on glycerol has also been observed.

A model is presented for the substrate specificity pattern of glycerol kinase of C. mycoderma. It postulates the involvement of three hydroxyl groups in the spatial distribution corresponding to the formation of L- α -glycerophosphate from glycerol. This requirement can be met by aldo- and ketotrioses in their respective hydrated forms.

The pathway of glycerol formation in *S. cerevisiae* has also been studied. Evidence is shown of the existence of a NADH dependent enzymatic activity reducing triose phosphate to α -glycerophosphate which can roughly account for the glycerol production. A low ionic strength seems to be required for the activity of this enzyme. The α -glycerophosphatase is specific for the L form, the efficiency of α -glycerophosphatase on D- α -glycerophosphate is 1/30 of that on L- α -glycerophosphate. The concentration of the α -glycerophosphatase in yeasts is higher when grown on hexoses than when grown on non-sugar carbon sources.

Specificity of the Constitutive Hexose Transport in Yeast. C. F. Heredia, A. Sols, and G. DelaFuente.

A method has been developed based on the osmotic sensitivity of protoplasts, that permits turbidimetric estimation of the penetration of non-metabolizable compounds into yeast protoplasts. Using this method a wide variety of sugars and related compounds have been tested as presumptive substrates for the constitutive hexose transport system in baker's yeast. An integration of the results obtained by this method with those obtained by other approaches has led to the establishment of the structural requirements that a compound has to meet in order to be transported. The basic structural requirement seems to be met by a pyranose ring for glucose and a furanose ring for fructose. With compounds that can be regarded as structurally related to D-glucopyranose, there is a broad tolerance for modifications at carbons 1 and 2, and somewhat less so for modifications at carbon 3. Similar requirements, except for carbon 2, apply to compounds that can be regarded as structurally related to D-fructofuranose.

Physical diffusion of sugars, and related compounds, through the cell membrane of baker's yeast is very low, so that they only can enter into the cell at measurable rates by means of specific transport devices. The factor by which the entrance of glucose is increased over its physical diffusion, at concentrations in the millimolar range, is of the order of 10^6 .

XV Research Laboratories of the State Alcohol Monopoly (Alko). Helsinki, Finland.
Communicated by Dr. Heikki Suomalainen.

Heikki Suomalainen. The structure and function of the yeast cell. Aspects of yeast metabolism, a Guinness Symposium, Dublin 1965, ed. by A. K. Mills and H. Krebs, Blackwell Scientific Publications, Oxford-Edinburgh 1968, p. 1-31.

The paper summarizes a part of the work performed at the Research Laboratories of the Finnish State Alcohol Monopoly. Investigations on the permeability characteristics of yeast, the properties of the plasma membrane in dried yeast cells, as well as the location of some enzymes in the yeast cell, particularly in the cell wall and the mitochondria, are discussed. Further, studies regarding the release of some compounds from the yeast cell into the medium during preparation of protoplasts are reported. The role of yeast in formation of the aroma compounds of different alcoholic beverages, with regard to the influence of the yeast strain, is discussed, and the influence of biotin on the fatty acids syntheses of yeast is studied.

Heikki Suomalainen and Lalli Nykänen. The aroma composition of alcoholic beverages. XXXVI^e Congrès International de Chimie Industrielle, Bruxelles 1966, Compt. Rend., Vol. 3, 1967, p. 807-811 (published 1968).

The volatile aroma compounds of alcoholic beverages consist mainly of alcohols, fatty acids, esters and carbonyl compounds. According to gas chromatographic analysis the qualitative similarity in the aroma composition is very striking in different beverages produced from different sources. This holds true for beers produced from malt, for wines from grapes and berries, for brandies and cognacs from grapes, for rums from cane molasses as for whiskies from grain and malt. Comparing the aroma compounds of alcoholic beverages with those synthesized by yeast in nitrogen-free sugar fermentation, it has been shown that yeast and fermentation play a central role in producing the aroma substances. The yeast strain is certainly of great significance.

Lalli Nykänen, Erkki Puputti and Heikki Suomalainen. Volatile fatty acids in some brands of whisky, cognac and rum. J. Food Sci. 33, 88-92 (1968).

Gas chromatography was applied to eight different types of whisky, two of cognac, one of brandy, and four of rum to determine the relative proportions of volatile fatty acids; with the lower molecular acids as free acids, but upwards from caprylic acid as methyl esters. Acetic acid and the total amount of volatile acids were measured quantitatively. Rum contained the largest amount of volatile acids, 600 mg/L, while one of the brands of Scotch whisky contained the least, 90 mg/L. Acetic acid represented 40-95% of the total amount of volatile acids in the whisky; for cognac and brandy, the value was 50-75%, and for rum 75-90%. The relative amounts have been reported for 21 acids, with acetic acid excluded. Capric, caprylic and lauric acid were the main components in whisky, cognac and brandy. Of the beverages analyzed, rum contained the largest quantity of lower fatty acids, particularly propionic and butyric acid; the main component of Jamaican rum was propionic acid. The main components of the group of long-chain fatty acids were myristic, palmitic and palmitoleic acids. Scotch whisky contained equal amounts of palmitic and palmitoleic acid; palmitoleic acid regularly appeared in smaller amounts in the other beverages.

Lalli Nykänen, Erkki Puputti and Heikki Suomalainen. Composition of the aroma in some brands of whisky and rum analysed by customary methods and by gas chromatography. Kemian Teollisuus 25, 399-404 (1968).

The brands of whisky produced in Scotland, in the United States and on the Continent, and those of rum produced in the West-Indies, contain up to 7.5 g extract, up to 100 mg aldehydes and 2100 mg fusel alcohols per litre. The ester content of rum varied between 45 and 630 mg per litre. One kind of imitation whisky has been found to contain fusel alcohols in exceptional proportions, 40% of isobutanol, 25% of n-propanol, 23% of isoamyl alcohol and 12% of opt.act.amyl alcohol. The fusel alcohols of the other brands of whisky mainly consist of isoamyl alcohol, along with isobutanol, opt.act.amyl alcohol and n-propanol in descending order. Moreover, the fusel oil of rum contains small amounts of sec-butanol and 2-pentanol.

Analyses have been made of the composition of the aroma fraction of two brands of South-European imitation whisky, and two brands of West-Indian white rum differing in strength. Twenty-four aroma components were found in the whisky, and fifteen in the rum; isoamyl alcohol was the main component in all

the samples. Both brands of whisky, as well as the heavy-flavour rum, additionally contained considerable amounts of ethyl acetate, isoamyl acetate, ethyl caprylate and ethyl caprate, the whisky had a further content of comparable amounts of ethyl laurate. The light-flavor brand of whisky had an abundance of high-volatile compounds present, while considerable quantities of long-chain fatty acid esters were found in the heavier type of whisky. The heavier type of rum contained larger amounts of esters, with the exception of ethyl acetate, than did the light rum.

H. Suomalainen and P. Ronkainen. Aroma components and their formation in beer. M B A A Technical Quarterly 5, 119-127 (1968).

The literature concerning the most important aroma components of beer and their formation is reviewed, and it is evident that little is known about the contributions of the yeast to the aroma of alcoholic beverages. Our own investigations show that the exceptional taste nuance of a Finnish lager beer is due to an unusually high content of fusel alcohols and isoamyl acetate, produced by a particular strain of brewer's bottom yeast. Gas chromatography reveals that the same aroma compounds are present in distillates of whisky, brandy, and nitrogen-free sugar fermentations. The central role of yeast in the formation of aroma components has been confirmed by the results of a comparison of the aroma composition of a berry wine of the sherry type with that of original Spanish sherries. Only minor differences are discernible in the composition of alcohols, esters, and acids in the beverages produced from different raw materials. However, when yeast is cultured aerobically in a medium, in which half of the ammonia nitrogen has been replaced either by L(+) valine-, L(+) leucine-, or L(+) isoleucine-, it is found that the raw material also influences the composition of the fermented product, although to a lesser extent. Isobutyric acid predominates when valine is added; isovaleric acid predominates when leucine is added, and α -methylbutyric acid predominates when isoleucine is added.

T. Nurminen, E. Oura and H. Suomalainen. Properties of the plasma membrane preparation obtained from the cell wall fraction of yeast. FEBS, Fifth Meeting, Prague 1968, Abstr. Commun. p. 115.

A carefully isolated cell wall preparation of yeast contains fragments of the plasma membrane. The bulk of the carbohydrates of the cell wall can be removed by enzymatic digestion and, after that, the fraction in which the plasma membrane is enriched can be isolated by centrifugation. The membrane sediment, thus obtained, contains as main components proteins and lipids, and further, an appreciable amount of carbohydrates. An activity of Mg^{++} -dependent ATPase is present, while, by digestion, the saccharase and the acid phosphatase of the cell wall fraction almost completely go into solution. It is characteristic that the cell wall fraction lacks short-chain fatty acids and instead contains principally the fatty acids $C_{18:1}$, $C_{16:1}$, and C_{16} , of which the $C_{16:1}$ predominate in the original cell wall fraction and $C_{18:1}$ in the digested preparation. The lipid phosphorus remains in the membrane preparation more readily than do the other phosphorus compounds. In addition to neutral lipids including sterols, the lipids contain phosphatidyl ethanolamine, phosphatidyl choline and phosphatidyl inositol+phosphatidyl serine.

Heikki Suomalainen and Lalli Nykänen. Methods applied in studies on the aroma composition of alcoholic beverages. Wallerstein Labs. Commun. 31, 5-15 (1968).

A review is given of analytical methods for the determination of the volatile aroma components in alcoholic beverages. Paper and thin layer chromatographic methods have been adapted to the identification of carbonyl compounds as 2,4-dinitrophenylhydrazones. In the main, gas chromatographic assays have been used for the analysis of the aroma fraction, containing alcohols, esters, and acids, separated from the alcoholic beverages by extraction.

Heikki Suomalainen, Dagmar Vraná and Erkki Oura. Changes in the morphology and nucleic acid content of baker's yeast cells during industrial production. Suomen Kemistilehti 41B, 284-288 (1968).

During the industrial production of baker's yeast, the size of the yeast cell diminishes on transfer to the aerobic yeast stages. The cells of the semiaerobically cultured, nutrient-rich yeast stages contain over three times as much dry matter as the yeast cells of the commercial stage. The relative age of the cells was determined from the number of budding scars. Scar-free cells predominated in the anaerobic yeast stage to the extent of 74%, while only 29% of the cells in the commercial yeast stage were scar-free. The amount of ribonucleic acid per unit weight of yeast decreased on transfer to the commercial stage, while the amount of deoxyribonucleic acid increased or, when calculated on the number of cells, remained constant. The synthesis of deoxyribonucleic acid was more rapid in the cells of anaerobically grown yeast stages than in those grown aerobically: thymidine labelled with tritium was more readily incorporated in the former than in the latter cells. The cells of the anaerobic yeast stage did not react to the "Division-Inducing Factor" isolated from Candida utilis, although this factor caused a 20% increase in the number of aerobically grown yeast cells.

XVI The University of New South Wales, School of Biological Technology, Box 1, Post Office, Kensington, 2033, Australia. Communicated by Dr. Pamela A. D. Rickard.

Abstract of paper entitled "The Response of Microorganisms to Controlled Concentrations of Oxygen and Glucose -- (1) Candida utilis" Submitted to Biotech. and Bioeng. F. J. Moss, Pamela A. D. Rickard, G. Beech and F. E. Bush.

SUMMARY

Dissolved oxygen and glucose concentration have been independently maintained at various concentrations for extended periods during growth of Candida utilis in continuous culture. Simultaneous observations of cytochromes A, B and C, oxygen uptake, CO₂ and ethanol output, growth rate, acid production, and rate of uptake of glucose have been made during steady states at various levels of oxygen and glucose. There is an inverse relationship between dissolved oxygen and cytochrome and between glucose concentration and cytochrome.

Studies of the transient state following a step change from high to low dissolved oxygen show that there is a lag of about 10 hr. during which there

is no change in the above parameters. This is followed by rapid oscillatory changes in cytochrome content and a change to a more fermentative metabolism.

CONCLUSIONS

1. The cytochrome content of C. utilis is dependent on the concentration of both oxygen and glucose. When the feed rate of glucose was growth-limiting, the concentration of cytochromes A, B and C attained a maximum value when dissolved oxygen was less than 0.1 μM . Excess glucose suppressed cytochrome particularly when dissolved oxygen was growth-limiting.
2. There is not a simple relationship between dissolved oxygen, cytochrome content, oxygen uptake, CO_2 output, growth rate, ethanol production or the production of organic acids.
3. Oxygen uptake increased as did dissolved oxygen when the feed rate of glucose was not growth-limiting. When glucose was growth-limiting $Q\text{O}_2$ was generally much less than when it was not limiting. In the case of non-limiting glucose, very high values of $Q\text{O}_2$ were recorded when dissolved oxygen was near to air saturation. $Q\text{O}_2$ was greater than expected in terms of growth and $Q\text{CO}_2$. It is possible that oxygen is taken up by oxygenases particularly when dissolved oxygen is near to saturation.
4. C. utilis grows very slowly anaerobically. It seems to be poorly endowed with the enzymes of the EMP pathway on the evidence of ethanol production. When glucose was in excess, ethanol was produced at all levels of dissolved oxygen.
5. The uptake rate of glucose increased as a direct function of dissolved oxygen when the glucose feed rate was not rate-limiting. The distribution of ingested glucose carbon to cell substance, glycolysis, non-glycolytic pathways and excreted organic acids was more dependent on the degree of growth limitation by the glucose feed rate than on dissolved oxygen.
6. When a step change from high to low oxygen is imposed, a smooth transition to a new steady state does not follow. The transient state is marked firstly by a lag of many hours during which $Q\text{O}_2$ is the only parameter to show significant change. This period is followed by the onset of rapid and irregular oscillations in cytochrome content and a change to an anaerobic type of fermentation as the new steady state is approached. The length of the lag, type of oscillations and overall length of the transient period were dependent on the degree of oxygen limitation.

Abstract of paper entitled "The Carbon Monoxide-Reactive Haemoproteins of Yeast", submitted to *Biochimica et Biophysica Acta*, Bioenergetics Section. T. C. K. Mok, Pamela A. D. Rickard and F. J. Moss.

The yeasts Saccharomyces cerevisiae, Candida utilis and Saccharomyces carlsbergensis were shown to have two CO-binding haemoproteins in addition to cytochrome a_3 . These haemoproteins were separated by a method described. One, contained in the particulate fraction of the cells, was extracted by sodium deoxycholate and precipitated at 30% ammonium sulphate saturation. The visible spectrum was that of a B-type cytochrome, but it reacted with carbon monoxide to produce CO-difference transmission minima at 408 $\text{m}\mu$, 540 $\text{m}\mu$ and 572 $\text{m}\mu$. It did not form a complex with either cyanide or oxygen. The other

haemoprotein was separated from the non-particulate cellular fraction and precipitated at 65% ammonium sulphate saturation. Like the haemoglobin of blood, this haemoprotein can be oxygenated, reduced and oxidized and can react with cyanide and carbon monoxide. The CO-difference transmission minima are situated at 419 m μ , 532 m μ and 568 m μ .

CO-difference reflectance spectrophotometry of C. utilis cells was employed for semi-quantitative estimation of the carbon monoxide complex of cytochrome a₃ (CO-a₃) and the carbon monoxide complex of those haemoproteins with a 570 m μ CO-difference reflectance minimum (CO-Hp₅₇₀). The concentration of CO-a₃ and CO-Hp₅₇₀ varied with glucose concentration and culture age. Evidence is presented for contribution to the CO-Hp₅₇₀ of C. utilis by both cytochrome o and haemoglobin in high glucose cultures and by cytochrome o alone in low glucose cultures.

XVII The Australian Wine Research Institute, 230 East Terrace, Adelaide, South Australia. Communicated by Dr. B. C. Rankine.

Part of the microbiological work carried out by The Australian Wine Research Institute has been a critical study of the importance of yeasts in influencing the composition and quality of wines, and a review of this work has now been published (Vitis, 1968 7 22-49). This deals with the taxonomy of yeasts used in the primary fermentation of grape juice (mainly Saccharomyces species) and yeasts which may cause spoilage, and goes on to discuss the influence of yeast strain on various products of fermentation and the way in which they influence the composition and quality of wine. Examples of the fermentation products studied are ethanol and various higher alcohols; acetaldehyde, pyruvic acid and α -ketoglutaric acid, and their effect on binding of sulphur dioxide; hydrogen sulphide; and malic acid decomposed during fermentation. Aspects of yeast dominance in mixed culture, and identification of closely-related strains by various biochemical tests are also described.

I report with regret the death in August 1968 of J. C. M. Fornachon, the Foundation Director of The Australian Wine Research Institute. Fornachon was a distinguished microbiologist who made important contributions to wine microbiology in the fields of sherry yeasts and lactic acid bacteria. He was the author of two monographs and numerous papers, and was regarded as an authority on wine microbiology.

XVIII Brewing Industry Research Foundation, Nutfield, Redhill, Surrey, England. Communicated by Dr. A. H. Cook.

Much of our work in the Microbiological Laboratories is concerned with the properties of strains of Saccharomyces cerevisiae used in brewing. The behaviour in batch fermentation of 150 distinct strains has been assessed and the results reveal a very wide range of properties (R. J. Walkey and B. H. Kirsop, J. Inst. Brew., in the press). Arising from this work more detailed studies of particular aspects of behaviour have been undertaken. The cells of different strains vary in their tendency to form a yeast head, that is to concentrate in the foam above the fermenting liquid. These variations have been related to the readiness with which cells concentrate at the surface of gas bubbles evolved during fermentation. Variations in fermentation medium are also important in that they alter the persistence of the yeasty foam; if this collapses rapidly the quantity of suspending yeast remains high. (I. J. Dixon and B. H. Kirsop, J. Inst. Brew., in the press). Mr. K. Visuri is studying the gradual loss of the ability to ferment maltotriose which is a characteristic aspect of some yeasts during fermentation of brewery wort.

The study of the serological properties of yeast cells by an immunofluorescence procedure is continuing and offers promise of increasing our understanding of the nature of the yeast cell wall. (T. W. Cowland, J. Inst. Brew., 74(5), 457, 1968.

Turning to very different yeast studies, visual and "Coulter-Counter" observations using a range of brewing strains of *S. cerevisiae* have provided interesting insight into changes in the size of individual cells and changes in the degree of association between individual cells during normal fermentation procedures. It has been found that flocculent yeasts in particular develop unusually high proportions of cell-walls in relation to the mass of the whole cells. Leaving out of account numerous other yeast studies which have been concerned more with the over-all course of fermentations than with scientific aspects of yeasts as such, much interest attaches to observations of the apparent maltase content of brewing yeasts during fermentation. It seems that these contents fluctuate unpredictably and not in such a way as to support the idea that maltase is formed by an adaptive mechanism.

XIX Lehrstuhl für Mikrobiologie der Technischen Universität Berlin und Forschungsinstitut für Mikrobiologie im Institut für Gärungsgewerbe und Biotechnologie, Seestraße 13, 1 Berlin 65 (West). Communicated by Prof. Dr. S. Windisch.

Since the last contribution the report about the first German symposium concerning "working methods and actual results of technical microbiology" (cf. Yeast Letters XV, Nr. 1, p. 7/8) has appeared as Supplementheft 2, 1967, of the Zentralblatt f. Bakt. I Orig., Verlag Fischer, Stuttgart. It contains 30 papers.

The following publications have appeared:

- S. Windisch: Possibilities and limits of the quantitative microbiological analysis of air with Bronn's impinger. Zbl.f.Bakt.I Orig. 198, 89-91, 1965
- S. Windisch: Is there a need for collections of microorganisms? Zbl.f.Bakt.I Orig., Supplementheft 2, 233-236, 1967
- S. Windisch: G. Boerner, Sigrid Janssen u. Ingrid Stern: Experiments to control *Saccharomyces* yeasts for requirements of vitamins and to breed anaerobic strains. Branntweinwirtschaft 107, 429-443, 1967

H.-J. Keßler has published his thesis "Über einige, das Wachstum von Hefen bestimmende Faktoren" in Zbl.f.Bakt. II 121, 129-178, 1967. This paper contains a lot of new results about growth parameters of yeasts.

XX International Meetings.

1. The First International Conference on Culture Collections (ICCC) was held in Tokyo, "Takanawa Prince Hotel", October 7 - 12, 1968. Communicated by Dr. A. Kockova-Kratochvilova.

The ICCC was held under sponsorship of the Japanese Government, UNESCO, ICRO-panel for applied microbiology, and the IAMS. The aim of the ICCC was to exchange information on existing culture collections of the world and

to review the present conditions in the field concerned and to contribute further to the development of an international network of culture collections.

The hospitality of the Organizing Committee enabled me to participate in this very important and interesting conference, to see Japanese working activities and life in Tokyo, the greatest and most imposing city I have seen, to visit many scientific institutes in Tokyo and Osaka and to meet with all my Japanese friends.

The working programme of ICCC was divided into three parts:

- 1) Plenary session, where the conditions of international and regional collections were discussed.
- 2) Special meeting, where the emphasis was on new ideas.
- 3) Symposia, which brought forth the scientific research results connected with culture collections.

More than 332 participants from 27 countries of the whole world were present and 84 lectures were presented. The operational course of the ICCC resulted in some important conclusions: The recommendation of the establishment of an International Federation for culture collections and a committee was appointed for preparing a draft constitution for submission to the 10th International Congress of Microbiology, in 1970. ICCC recommended that the next conference be held in Czechoslovakia in 1972, that a special technical course in the methods of preservation of cultures be organized in 1970. The ICCC further recommended establishment of reference laboratories, an International Center for characterization of strains of microorganisms and of an International Center for Information and a handbook of methods for conservation of cultures has to be published.

The Organizing Committee of the ICCC did a perfect job, the course of the conference was faultless, the visits to research and university institutes and the excursions to Osaka, Nara, Kyoto, Hakone and Kamakura were excellent.

2. Your editor attended the Second International Symposium on Yeast Protoplasts held on Aug. 19-24, 1968 in Brno, Czechoslovakia. The meeting was organized by a committee under the chairmanship of Prof. O. Nečas, J. E. Purkyne University, Brno.

Approximately 50 scientists from many countries attended this very well organized meeting. Thirty-five lectures were presented which will be published in the form of "Proceedings of the Symposium" at the end of 1969. In spite of the depressing circumstances surrounding the meeting in the form of the occupation of Czechoslovakia by foreign armies, the International Symposium was successfully held and completed. Under these trying conditions the Organizing Committee deserves high praise for its efforts to continue this long planned symposium under such adverse conditions. I am sure that I speak for all participants in complimenting Prof. Nečas and his committee for their splendid efforts.

3. Prof. Susumu Nagai, Biological Laboratories, National Women's University, Nara, Japan, has sent your editor a copy of "Abstracts of Presentations

at the Yeast Genetics Conference", 2-5 Sept. 1968, Osaka, Japan. The abstracts cover papers given by twenty-three yeast geneticists and their collaborators in the form of a 47 page proceedings.

XXI Brief News Items

1. Instituto Nacional de Investigaciones Agronomicas, Seccion de Bioquimica, Madrid, Spain. Prof. J. Santa Maria writes that the following articles have been published recently.

J. Santa Maria: "Saccharomyces hispanica, nov. spec. nueva especie de levadura de "flor". ("Saccharomyces hispanica, nov. spec., a new "flor" yeast"). Boletin Inst. Nac. Invest. Agronomicas XXVIII, 21-32, 1968.

A. Rodriguez: "Flora zimogena de las mieles espanolas". ("Yeast flora of spanish honey"). Boletin Inst. Nac. Invest. Agronomicas XXVIII, 33-42, 1968.

Current research interests include:

- 1 - Studies on yeast taxonomy: the main problems being investigated are:
 - a) characteristics delimiting the genus Kluyveromyces;
 - b) a study on the typical spanish "flor" yeasts.

- 2 - Mating types in different strains of the genus Saccharomyces.

2. Department of Microbiology, Miami University, Oxford, Ohio 45056. Dr. J. K. Bhattacharjee informs the readers that the following reports, related to the lysine biosynthesis in yeast, appeared this year from the Murray Strassman Laboratory, A. E. Medical Center, Philadelphia (author's previous address).

1. Accumulation of alpha-ketoglutaric acid in yeast mutants requiring lysine. J. K. Bhattacharjee, A. F. Tucci, and M. Strassman. Arch. Biochem. Biophys., 123:235 (1968).

2. Identification of malic acid from yeast. J. K. Bhattacharjee, M. E. Maragoudakis, and M. Strassman, J. Bacteriol., 95:494 (1968).

3. Relationship of glutaric acid to lysine biosynthesis. J. K. Bhattacharjee, A. F. Tucci, and L. N. Ceci. Federation Proc., 27:794 (1968).

4. Genetic basis for the homocitrate pathway for lysine biosynthesis in yeast. Proc. XIIth Int. Nat. Cong. Genet., Tokyo (Japan) (1968).

5. Preparation of homocitric, homoaconitic, and homoisocitric acids. A. F. Tucci, L. N. Ceci, and J. K. Bhattacharjee. Methods in Enz. (in press) (1968).

3. Dr. M. C. Pignal, Université De Lyon, Laboratoire de Biologie Végétale, 43, Boulevard Du 11 Novembre 1918, 69 Villeurbanne, France, writes: Since the last issue of the Yeast News Letter, the following articles have appeared:

F. Abadie Assimilation des nitrites et de composés organiques par quelques levures et organismes levuriformes. Annales de l'Institut Pasteur 115: 197. 211 1968.

F. Abadie Essai d'utilisation de quelques composés azotés organiques dans la systématique des genres Pichia, Debaryomyces et Saccharomyces (levures ascoporées). Revue de Mycologie 32 (4): 278. 286. 1967.

R. Montrocher et F. Abadie Essai d'utilisation taxonomique de l'assimilation azotée de composés organiques dans le genre Candida (levures Cryptococcaceae). Revue de Mycologie 32 (4): 287. 299 1967.

Several other articles are in press.

4. Illinois Institute of Technology, Department of Biology, Chicago, 60616.

Two papers have been recently published from work in our laboratories. These are:

L. R. Hedrick and P. D. Dupont. 1968. The utilization of L-amino acids as carbon source by yeasts of the genera Hansenula and Trichosporon. Antonie van Leeuwenhoek 34:465-473.

L. R. Hedrick and P. D. Dupont. Two new yeasts: Trichosporon aquatile and Trichosporon erienze spp. n. Antonie van Leeuwenhoek 34:474-482.

Ken Shieh, who has been studying the energy requirements for the uptake and utilization of glutamate by Hansenula subpelliculosa, has completed his work for the Ph.D. and is working in the enzyme laboratories at Anheuser Busch, St. Louis, Missouri.

Work is continuing upon the yeasts associated with fresh water from lakes, streams, and glaciers, and from aquatic environments with varying degrees of pollution.

L. R. Hedrick

5. MacDonald College of McGill University, Department of Microbiology, Que. Canada.

Dr. Blackwood relinquished the Chairmanship of the Dept. of Microbiology, MacDonald College in May 1968 to devote full time to research and teaching. He is presently on a nine month sabbatical leave at the University of Bordeaux, Bordeaux, France, where he is engaged in Research on a acetic acid fermentation.

He was made a Fellow of the Royal Society of Canada in 1968. At the present time he has a graduate student working full time on the problem of "yeast species in the St. Lawrence River".

6. New England Medical Center Hospitals, 171 Harrison Avenue, Boston, Mass. 02111

The following paper has just been published: "Routine Identification of Yeasts with the Aid of Molybdate-Agar Medium", Applied Microbiology, Oct. 1968, p. 1503-1506. The following is the summary:

"A large number of yeasts, including a variety of species other than Candida albicans, were isolated from clinical specimens. C. tropicalis and Torulopsis glabrata were each found one-third as frequently as C. albicans. A schema is presented which made possible, by simple procedures, the

identification of the great majority of the isolated yeasts. Preliminary isolation and differentiation was aided by the use of molybdate-agar medium. The use of the schema by diagnostic bacteriological laboratories is discussed."

Charles M. Bump, Chief, Bacteriology Lab

7. Univ. Doz. Dr. B. Gedek of the Institut für Mikrobiologie und Infektionskrankheiten der Tiere, der Ludwig-Maximilians, Universität, München, Germany writes: I have changed my last name from Mehnert to Gedek and I have published a monography: Gedek, Brigitte; Hefen als Krankheitserreger bei Tieren. It was published by the VEB Gustav Fischer Verlag in Jena, D.D.R. Autumn 1968.
8. Department of Food Science and Technology, University of California, Davis, Calif. 95616.

Publications:

Ahmed T. H. Abd El-Al and H. J. Phaff. Exo- β -glucanases in yeast. Biochem. Jour. 109:347-60, 1968.

Sally A. Meyer and H. J. Phaff. Deoxyribonucleic acid base composition in yeast. J. Bacteriol. 97: January 1969.

J. I. Pitt and M. W. Miller. Sporulation in Candida pulcherrima, Candida reukauffii and Chlamydozoma species: Their relationships with Metschnikowia. Mycologia 60:663-685, 1968.

Dr. A. Martini from the University of Perugia, Italy, has joined our group to study DNA homologies and base ratios of yeasts as a taxonomic aid under a NATO fellowship.

Drs. Miller and Phaff accompanied by Dr. Yoneyama and Mr. Soneda of Japan returned Aug. 1, 1968 from a yeast collecting trip through Alaska, the Yukon Territory, British Columbia, Washington and Oregon. About 700 strains were isolated from tree exudates and related habitats and these cultures are in the process of identification.

Dr. Phaff and Dr. E. J. Buecher presented a paper entitled "On the Cell Wall Composition of Saccharomycopsis guttulata (Robin) Schiöningg" during the second international symposium on yeast protoplasts, held at Brno, Czechoslovakia in August 1968.

9. Professor H. Kaspar von Meyenburg, Dept. of Microbiology, Federal Institute of Technology, 8006 Zürich, Switzerland, reports that the article "Energetics of the budding cycle of Saccharomyces cerevisiae during glucose limited aerobic growth" will be published in the Proceedings of the 4th International Symposium on continuous cultivation of microorganisms, Prague 1968.

"In the present paper a calculation of the yield on energy Y_{ATP} for different P/O ratios is performed based on the measurement of glucose catabolism of baker's yeast by the analysis of metabolized gases (FIECHTER & v. MEYENBURG, in press) in function of the specific growth rate μ in aerobic continuous culture with glucose limitation (FIECHTER & v. MEYENBURG, 1966a,b; v. MEYENBURG, 1968). This serves to examine the proportionality

between growth and energy formation (constancy of Y_{ATP}) and to find the effective P/O ratio for intact growing cells of Saccharomyces cerevisiae."

10. National Research Council of Canada, Division of Biosciences, Ottawa 7, Canada. I shall be working with Donald Williamson at the National Institute for Medical Research, Mill Hill, London, for a year, beginning September 1, 1968.

Byron F. Johnson