

Y E A S T

A News Letter for Persons Interested in Yeast

November 1962

Volume XI, 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

	Page
Mrs. N. J. W. Kreger-van Rij, Delft, Holland	17
Juan Santa Maria, Madrid, Spain	18
I. Banno and T. Hasegawa, Osaka, Japan	18
Shoji Goto, Kofu, Japan	19
Akira Yuasa, Tokyo, Japan	19
A. G. Marr, Davis, California	20
A. H. Rose, Newcastle-on-Tyne, England	21
Colin H. Clarke, Edinburgh, Scotland	21
J. Oliver Lampen, New Brunswick, New Jersey	22
F. M. Clark, Urbana, Illinois	23
J. Wynants, Bruxelles, Belgium	24
John Kleyn, Seattle, Washington	24
A. L. Demain, Rahway, New Jersey	25
H. J. Phaff, Davis, California	25
K. Vas, Budapest	26
Brief News Items	28

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 1963. A contribution of \$0.50 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 Editors

I. Centraalbureau voor Schimmelcultures, Yeast Division, Julianalaan 67A,
Delft, Holland, Communicated by Mrs. N. J. W. Kreger-van Rij.

The following new species, for which a description was published,
have been received by the C.B.S.

Bullera teugae Phaff et do Carmo-Sousa
H. J. Phaff & L. do Carmo-Sousa,
Antonie van Leeuwenhoek, 28: 193, 1962.

Candida oregonensis Phaff et do Carmo-Sousa
H. J. Phaff & L. do Carmo-Sousa,
Antonie van Leeuwenhoek, 28: 193, 1962.

Cryptococcus skinneri Phaff et do Carmo-Sousa
H. J. Phaff & L. do Carmo-Sousa
Antonie van Leeuwenhoek, 28: 193, 1962.

Paratorulopsis banhegyii Galgóczy et Novák
J. Galgóczy and E. K. Novák, Acta Microbiologica
Academiae Scientiarum Hungaricae, 9 (1): 77, 1962.

Pichia saitoi Kodama, Kyono et Kodama
K. Kodama, T. Kyono and S. Kodama,
J. Gen. Appl. Microbiol., 8: 52, 1962.

Saccharomyces hienipiensis Santa Maria
J. Santa Maria, J. gen. Microbiol., 28: 375, 1962.

Schwanniomyces persoonii v.d. Walt
J. P. van der Walt, Antonie van Leeuwenhoek, 28: 81, 1962.

Sporobolomyces singularis Phaff et do Carmo-Sousa
H. J. Phaff & L. do Carmo-Sousa, Antonie van Leeuwenhoek,
28: 193, 1962.

Torulopsis burgeffiana Benda
I. Benda, Antonie van Leeuwenhoek, 28: 208, 1962.

Torulopsis haemulonii van Uden et Kolipinski
N. van Uden and M. C. Kolipinski, Antonie van Leeuwenhoek,
28: 78, 1962.

Trichosporon atlanticum Siepmann
R. Siepmann and W. Höhnk, Veröffentlichungen des
Instituts für Meeresforschung in Bremerhaven, 8: 79, 1962.

Trichosporon maritimum Siepmann
R. Siepmann und W. Höhnk, Veröffentlichungen des
Instituts für Meeresforschung in Bremerhaven, 8: 79, 1962.

Trichosporon piscium Siepmann
R. Siepmann und W. Höhnk, Veröffentlichungen des
Instituts für Meeresforschung in Bremerhaven, 8: 79, 1962.

II. Instituto Nacional de Investigaciones Agronomicas, Seccion de Bioquimica, Madrid, Spain. Communicated by Dr, Juan Santa Maria.

Since the last publication of the Yeast News Letter the following papers have been published:

"Saccharomyces hienipiensis, a New Melibiose-fermenting Yeast, Unable to Assimilate Raffinose". J. gen. Microbiol. (1962), 28, 375-384.

"Fermentation of Saccharose by Alpha-Glucosidase". Nature, Vol. 195, No. 4847, pp. 1201-1202, September 22, 1962.

I would like to announce the forthcoming publication of the following new species:

Torulopsis salmanticensis nov. spec.

Growth on malt extract: after 3 days at 25°C, cells are round and oval, single or in pairs. (1,9-4,5) x (2,0-6,5)μ. A sediment is formed.

Slide culture: no pseudomycelium is formed.

Sporulation: spores are not formed.

Fermentation: Glucose +, galactose +, saccharose +, maltose +, lactose -, melibiose +, trehalose +, raffinose +, melezitose +.

Assimilation: glucose, galactose, L-sorbose, maltose, saccharose, cellobiose, trehalose, melibiose, raffinose, melezitose, inulin, D-xylose, D-arabinose, D-glucosamine hydrochloride, ethyl alcohol, adonitol, D-mannitol, D-sorbitol, alpha-methyl-D-glucoside, salicin, pyruvic acid, DL-lactic acid, succinic acid, citric acid (weakly) are assimilated. In some of these tubes films are formed.

Assimilation of KNO₃: negative.

Growth on a vitamin free medium: none.

Growth in the osmotic pressure medium: positive, after 7 days at 25°C.

Growth at 37°C: negative.

III. Institute for Fermentation, Juso-Nishino-cho, Higashiyodogawa-ku, Osaka, Japan. Communicated by Dr, I. Banno and Dr. T. Hasegawa.

The work on Rhodotorula yeasts has continued in our laboratory. Some strains of Rhodotorula were considered to be in haplophase from the nature of a survival curve after X-ray irradiation. An attempt to reveal the sexual interaction between various strains in Rhodotorula glutinis was made. When yeast cell types of auxotrophic double mutants of Rhodotorula gracilis and Rh. koishikawensis were mixed together in glucose-yeast extract-salt medium, prototrophic colonies appeared on minimal medium only from the mixed culture. Furthermore, fusing cells in pair were observed among the mixed cells by microscopic examination. It was, therefore, manifest that prototrophic colonies resulted from an interaction of cells of the two parental strains. It is noteworthy that the prototrophic growth consists of mycelium with clamp connections at septa. The fact that the mycelium has clamp connections suggests that the mycelial growth of this organism probably corresponds to secondary, dikaryon mycelium observed generally in Basidiomycetes.

A communication of the fact will be published presently. In addition to the investigation of the life cycle and sexuality in this organism, the search for more compatible pairs among various strains of *Rhodotorula* is now in progress.

IV. R. Institute of Fermentation, Yamanashi University, Kofu, Japan,
Communicated by Dr. Shoji Goto.

Identification of Film Yeasts Maintained in Japan

Part 1 Genus Pichia

Thirty-nine strains of film yeasts, whose majority belonged to the genus Pichia, which had been maintained since 1954 in Japan, were taxonomically studied by the system of Lodder and Kreger-van Rij. Certain corrections in nomenclature were found necessary.

Both of the groups, Pichia miso and P. sake, were found to be the same species, and so they were named Pichia miso Mogi according to the international rules of nomenclature. Pichia rosa (P-16) was recognized as a new species of *Rhodotorula*, and was named Rh. rosa (Nishiwaki) nov. comb. Pichia polymorpha (P-15) and Zygopichia chevalieri var. fermentati (P-13) were respectively identified with Hansenula anomala and Saccharomyces cerevisiae var. ellipsoideus. Two unknown yeasts were respectively identified with Candida krusei (P-45) and Candida tropicalis (P-47).

Part 2 Genus Hansenula

Thirty-seven strains of film yeasts, whose majority belonged to the genus Hansenula, maintained in Japan were taxonomically studied by the system of Lodder and Kreger-van Rij.

Hansenula miso reported by Dr. Mogi was found to be the same new species as described in the original report. It is proposed to consider Hansenula miso var. octosporus and H. miso var. B as new varieties of H. miso, H. anomala var. productiva as a new variety of H. anomala and H. schneegii var. kinshii as a new variety of H. schneegii. It is also proposed that a non-sporulating yeast (Hansenula sp. S-24) should be recognized as a new variety of Candida parapsilosis and it was named Candida parapsilosis var. hokkaii.

Bull. R. Inst. of Fermentation, Yamanashi Univ. (Japan) 9 (1962),
in press.

V. Department of Biology, College of General Education, University of Tokyo,
Komaba, Meguroku, Tokyo. Communicated by Prof. Akira Yuasa.

The seminar of yeast-cell studies

The seminar of yeast-cell studies in Tokyo has about 80 members and has a meeting once a month. It has also a big meeting once a year at which about one hundred investigators attend and discuss various problems on yeast.

Among the members of this seminar there are professors or assistant professors of universities, investigators of institutes and investigators

of brewing companies. They are biochemists, cytologists, physiologists, taxonomists or geneticists. At this seminar one member talks about his work on yeast and the attending members discuss this problem with him. Sometimes an investigator of another field is invited to talk at this seminar about his studies, for example, about cytology of microorganisms, membrane-permeability of fungi and so on.

The aim of this seminar is to study yeast cells from various points of views of biology and to contribute to the complete understanding of yeast cells.

The present chairman of this seminar is Dr. Akira Yuasa, Department of Biology, College of General Education, University of Tokyo, Komaba, Meguroku, Tokyo, Japan, who invites correspondence from interested scientists elsewhere.

The titles of the speeches which were given at this seminar are, for example, as follows:

- Akira Yuasa: Cytological studies on yeast cells.
- Tsuyoshi Tsuchiya: Antigen and anti-body reaction and its application.
- Hiroshi Onishi: Resistance of yeast cells to salt.
- Noboru Sando: Spore-formation in yeasts.
- Yoshiaki Yoneta: On the structure of nuclei in yeasts.
- Masahiro Takahashi: Vitamin and amino acid requirements of yeasts.
- Takeshi Tanaka: Ultra-violet ray mutation in yeasts.
- Yoichi Otaka: Structure and stability of ribosomes in yeasts.
- Kenji Tsukahara: On the Sake-yeasts.
- Yasushi Yamamoto: On the beer-yeasts.

The following list shows the subjects and speakers of the symposium which was held on Oct. 12, 1961 in Tokyo, under the auspices of "Seminar of Yeast-study".

1. Gyozo Terui and Tosihiro Enatsu: Anthranilic acid metabolism and tryptophan-biosynthesis of yeasts.
2. Tosiaki Takahashi: Linkage-group of yeasts.
3. Seno-Kanji and Jozi Ashida: Genetics of Cu-resistance in Saccharomyces cerevisiae.
4. Gen Kuraishi: Unbalanced growth of yeasts.
5. Akio Maeda: Structure and protein-synthesis of ribosomes.

VI. Department of Avian Medicine and Department of Bacteriology, University of California, Davis, California, U.S.A. Communicated by Dr. M. Shifrine and Dr. A. G. Marr.

The following is a summary of a paper submitted to the Journal of General Microbiology:

The Requirement of Fatty Acids by Pityrosporum ovale

SUMMARY

Fatty acids are required for the growth of Pityrosporum ovale (Bizzozero) Castellani et Chalmers. Myristate or palmitate satisfies this

requirement. Oleate increases the crop of cells in a medium containing limiting concentrations of myristate or palmitate. When myristate-1-¹⁴C was added to the medium, the cells of P. ovale contained palmitate, stearate, oleate, and linoleate with approximately the same molar specific radio-activity as myristate. Thus, P. ovale can synthesize both saturated and unsaturated fatty acids of higher molecular weight from myristate.

VII. Department of Bacteriology, King's College, University of Durham, Newcastle-on-Tyne, England. Communicated by Dr. A. H. Rose.

Mr. Bernard Dixon has been studying the role of biotin in the synthesis of carbamoyl phosphate ornithine carbamoyl transferase by Saccharomyces cerevisiae and, in September of this year, read a paper before the Society for General Microbiology embodying his main findings. On completing these studies, Mr. Dixon is planning to examine the effect of biotin deficiency on the structure of membranes, mitochondria and ribosomes in yeast. Mahboob Iqbal has joined the team working on biotin metabolism; he is on leave from the West Regional Laboratories of the Pakistan Council of Scientific and Industrial Research. His main research project is the biosynthesis of biotin.

Work has been resumed on the biochemistry of the psychrophilic habit in yeasts. Mrs. Lillian M. Evecon has already commenced an examination of the temperature relationships among a group of psychrophilic yeasts, which include Candida, Madsonia and Rhodotorula spp. Mr. Simon Stanley collected material during a recent expedition to Labrador, and is presently examining this for psychrophilic micro-organisms. A psychrophilic strain of Rhodotorula was found in lake water from this region.

The following publications have appeared from this laboratory during the past year:

- Hagen, P-O. & Rose, A. H. Studies on the biochemical basis of the low maximum temperature in a psychrophilic cryptococcus. J. gen. Microbiol. 27, 89 (1962).
- Ahmad, F. & Rose, A. H. Effect of biotin-sparing substances on growth of biotin-deficient Saccharomyces cerevisiae and on the synthesis of nucleic acids and protein. J. gen. Microbiol. 28, 147 (1962).
- Ahmad, F. & Rose, A. H. The role of biotin in the regulation of enzyme synthesis in yeast. Arch. Biochem. Biophys. 97, 302 (1962).
- Rose, A. H. Effect of biotin deficiency on the stability of yeast RNA to extraction with aqueous butanol. Biochim. biophys. Acta, 61, 628 (1962).
- Rose, A. H. Temperature relationships among micro-organisms. Wallerstein Laboratories Communications, 25, 5 (1962).
- VIII. Institute of Animal Genetics, Induced Mutagenesis Group, West Mains Road, Edinburgh 9, Scotland. Communicated by Dr. Colin H. Clarke.

Present work with Schizosaccharomyces pombe is aimed at the further analysis of the inhibitory effect of L-methionine in the plating medium on

the phenotypic expression of spontaneous and HNO₂- or U.V.-induced adenine-independent reverse mutations.

Genetic analyses are being made of revertants induced by different agents from ten different *adn-1* mutants. Special tests made with revertants of a temperature sensitive *adn-1* auxotroph have failed to show the presence of intra-genic suppressor mutations.

Recently I have been able to pay a most useful visit for two months to Professor U. Leupold's department in Zurich, and to take part in a symposium on *S. pombe* held there.

Publications

Clarke, C. H. & Bond, G. "Induced reversions of adenine-1 mutants in *Schizosaccharomyces pombe*" (abstract) *Heredity*, in press.

Clarke, C. H. "A case of mutagen specificity attributable to a plating medium effect." *Z.f. Vererb.-Lehre*, in press.

Clarke, C. H. "A possible correlation between reverse mutation and Complementation." *Experientia*, in press.

IX. Institute of Microbiology, Rutgers, The State University, New Brunswick, New Jersey. Communicated by Dr. J. Oliver Lampen.

During the last year the following papers have been published or accepted for publication from our laboratory:

1. Lampen, J. O., Intermediary metabolism of fungi as revealed by drug reaction. In *Fungi & Fungus Diseases*, Symposium II of the N. Y. Acad. Med., Sect. of Microbiol., Springfield, Ill. Charles C. Thomas (1962) p. 102.

Essentially a review of our recent studies on the mechanism of action of polyene antifungals.

2. Shockman, G. D., and Lampen, J. O. Inhibition by antibiotics of the growth of bacterial and yeast protoplasts. *J. Bacteriol.* 84: 508 (1962).
3. Lampen, J. O., P. M. Arnow, Z. Borowska and A. Laskin. Location and role of sterol at nystatin-binding sites. *J. Bacteriol.* (December 1962).

Polyenes are bound almost instantaneously by protoplast membranes or cell walls from log-phase *S. cerevisiae* strain LK2G12, whereas the original cells still bind relatively slowly especially at 0°. The characteristics of binding by intact cells probably represent the requirements for either transport of the polyene to the binding site or "activation" of the site. The membranes contain unesterified ergosterol and this is concluded to be the critical site of polyene binding. Walls contain bound sterol which may also bind with nystatin; however, binding to the wall is not an essential feature of polyene action since protoplast growth and metabolism are sensitive (ref. 2).

4. Lampen, J. O., and P. M. Arnow. Differences in action of large and small polyene antifungal antibiotics. Bulletin of the Res. Council of Israel (March 1963).

The action of large (C₄₆₋₄₇) polyenes on yeast glycolysis is reversible by K⁺ and NH₄⁺ whereas inhibition by small (C₃₄₋₃₇) polyenes is not. The greater tendency of the small polyenes to produce protoplast lysis and "activation" of membrane supports the concept that the small polyenes produce more drastic damage to the cell membrane than do the large molecules. The structural basis for this difference in action cannot at present be defined.

5. Sutton, D. D., and Lampen, J. O. Localization of sucrose and maltose fermenting systems. Biochim. et Biophys. Acta 58: 294 (1962).
7. McLellan, Wm. L., and Lampen, J. O. The acid phosphatase of yeast: Localization and secretion by protoplasts. Biochim. et Biophys. Acta (in press).

Both acid and alkaline phosphatase in yeast were repressed by high concentrations of inorganic phosphate. The acid phosphatase was external to the cell membrane and released during preparation of protoplasts with snail enzyme. It was not, however, firmly attached to cell walls prepared by rupturing the organism. In the absence of the cell wall newly synthesized acid phosphatase (but not alkaline phosphatase) was secreted into the medium.

Dr. Mary Harsch has investigated the effect of N-acetylcandidin on K⁺ transport in yeast since this appears to be uniquely sensitive to polyene action. NAC-treated cells lose K⁺ to a K⁺-free medium but can still concentrate added K⁺ although less effectively than normal cells. K⁺ loss stops at 0°C, thus the antibiotic does not appear to produce true holes in the membrane. It is proposed that the combination of the polyene with the membrane causes sufficient damage to the transport systems that the cell can no longer concentrate essential low molecular weight substances effectively.

- X. University of Illinois, Urbana, Illinois. Communicated by Dr. F. M. Clark.

We have continued our work on the inositol compounds in the yeast Schizosaccharomyces pombe. Mr. Robert Kyndberg has isolated and at least partially identified one of these inositol fractions. The yeast cells after washing were dried with acetone and then exhaustively extracted with chloroform:methanol (2:1 v/v). The extract was fractionated chromatographically on glass plates containing a layer of silica gel G. The fractions found to contain bound inositol were eluted from the silica gel with chloroform-methanol. The inositol containing fraction eluted from this first test was again chromatogramed but resisted any further fractionation. Inositol, glycerol, inositol mono-phosphate, and glycerol mono-phosphate were identified by paper chromatography of acid hydrolysates of the fraction. Molar ratios closely approaching 1:1:2 were found respectively for inositol, phosphorous and acyl ester. Fatty acids present were determined by gas chromatographic techniques as palmitic and oleic acids. Percent weights

of inositol, phosphorous and C₁₆₋₁₈ fatty acids closely approached the theoretical yield for a simple monophospho-inositide containing an esterified palmitic and/or oleic acid. The fraction was considered to be either the theoretical inositide or a "family" of such inositides containing the determined fatty acids.

This appears to be only one of the forms of inositol found in this yeast. Cell residues after extraction as described above when hydrolyzed indicated other inositol compounds present in the cells that had not been extracted by the method used.

We have been doing some work in our laboratory on ascospore formation by yeast. Selected cultures of yeast grown under different conditions of nutrition are being tested for their ability to produce ascospores. We would like to know if nutritional factors determine the number of ascospore produced per cell.

XI. Brasserie St. Josse. 71-73 rue des Deux Tours, Bruxelles 3, Belgium.
Communicated by Mr. J. Wynants.

During the experiments on the storage of brewery yeast cultures by freeze-drying (from which some results have already been published, J. Wynants, 1962, J. Inst. Brewing 68, 350). A surprising number of abnormally small colonies were detected in the revitalized-lyophilized material. It was shown that isolated dwarf colonies had lost their respiration abilities, and that the cells were morphologically and physiologically similar to the Ephrussi respiration-deficient mutants.

We are of the opinion that the lyophil process induces the formation of mutants "Petites Colonies". It is based principally on the absolute increase in the number of dwarf colonies after lyophilization and it is also supported by a recent observation of the lower resistance shown by a freeze-dried "Petite".

XII. Sicks' Rainier Brewing Company, 3100 Airport Way South, Seattle 4, Washington. Communicated by Dr. John Kleyn.

Since the last issue of the Yeast News Letter the following papers have been published or prepared:

1. Published: J. Kleyn, R. Mildner, and W. Riggs. Yeast viability as determined by methylene blue staining. The Brewers Digest 37, 6, 42, 1962.
2. Presented at the annual meeting of the American Institute of Biological Sciences, Corvallis, Oregon, August, 1962: J. G. Kleyn and N. L. Vacano. Selection of superior brewer's yeast strains by a quaternary screening procedure.
3. In press:
 - a. Kleyn, J. G. Yeast dwarf cell formation. Journal of publication unannounced.

- b. Kleyn, J. G. and Vacano, N. L. Yeast dwarf cell formation as related to beer brewing. Journal of publication unannounced.

Our current biological research includes the following:

1. A study of certain growth and fermentation characteristics of 40 different yeast strains belonging to the genus Saccharomyces.
2. A study of biologically stable beer production by other methods than conventional pasteurization. As regards this research we are interested in obtaining as well as exchanging cultures of microorganisms (both bacteria and yeast) able to grow in previously fermented American lager beer.

XIII. Merck Sharp & Dohme, Research Laboratories, Rahway, New Jersey.
Communicated by Dr. A. L. Demain.

"Lysineless mutants of Saccharomyces cerevisiae, capable of growth with DL- α -amino adipic acid, were compared to those of Neurospora crassa as to their ability to use the D-isomer. The D- α -amino adipic acid was prepared by E. Bartnicki and D. Hendlin. It was found that the yeast mutants failed to grow with D- α -amino adipic acid. N. crassa, on the other hand, could use either the DL or the D form. The racemic mixture was used much more efficiently, however. Details will be published in the February issue of the Canadian Journal of Microbiology.

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

1. Dr. Michael Lewis is continuing work on a project dealing with the release of nitrogenous substances from brewer's yeast under various conditions of storage.

Nitrogen excretion was studied with an additional three strains of yeast. The ratio of nucleotides to amino acids was the same in the excreted fraction as inside the cells. The size of free internal pools of amino acids and nucleotides was shown to be strongly influenced by the growth conditions. Rate and extent of leaching of cell contents was partly determined by yeast strain and partly by physical conditions of storage. The sources of the excreted nitrogen are the free pools in the cytoplasm. Addition of glucose to a storage medium causes rapid reabsorption of amino acids, whereas nucleotides continue to be excreted. This reabsorption is coupled with synthesis of amino acids to protein and growth results. Accelerated excretion of nucleotides in the presence of glucose results in a correspondingly lowering of the nucleotide pool level. Shock excretion and the effect of cations and ethanol were studied also. Shock excretion is defined as the sudden release of amino acids from washed yeast upon exposure of the yeast to solutions of certain sugars. If the sugar can serve as an energy source the amino acid excretion is followed by a rapid reabsorption into the cell.

2. Dr. J. F. T. Spencer is studying the taxonomy and ecology of yeasts isolated from a number of natural sources.

Yeasts and yeast-like organisms were isolated from wild flowers collected in Saskatoon (Saskatchewan), Fort Smith (North West Territory) and Davis, California. Yeasts were also obtained from brine shrimp (saline Canadian lakes) and from various streams and lakes in Northern California. The flowers yielded mainly species of Cryptococcus, Rhodotorula, Torulopsis, Candida and Pullularia. Some species of Saccharomyces and Sporobolomyces were found also. The brine shrimp yielded a possibly new species of the recently described and rare genus Metchnikowia. One, as yet unidentified, species of yeast from flowers near Davis appears to produce, under certain conditions of growth, crystals of a glycolipid. Because of the many strains isolated of the genus Cryptococcus, it appears possible to improve the systematics of the species in this complex genus.

3. The project dealing with yeasts isolated from *Drosophila* flies collected in a Sacramento Valley orchard and in a nearby hill area was completed and is being prepared for publication.

4. Mr. H. Tanaka will be completing his work on yeast cell walls during early spring. A preliminary summary of his work follows.

A strain of Bacillus circulans, which produced enzymes hydrolytic towards baker's yeast walls, has been isolated from soil. The organism produces extensive clear zones around colonies growing on agar medium with baker's yeast cell walls as the carbon source. Baker's yeast walls were prepared by grinding compressed yeast with glass beads (average diameter 120 μ) in a colloid mill, yielding 38 g of dry walls per kg of compressed yeast. Enzyme excretion was induced by baker's yeast walls and was much greater in aerated cultures than in standing cultures. An assay procedure was developed with soluble laminarin as the substrate, measuring the rate of increase of aldehyde groups by alkaline hypoiodite. Laminarin, which contains mainly β -1-3 linkages, was hydrolyzed rapidly by a random mechanism to glucose, laminaribiose and laminaritriose. The last oligosaccharide was slowly hydrolyzed to the dimer and glucose. Laminaritetraose was split into glucose and the trisaccharide. The enzyme preparation also hydrolyzed pustulan, a polyglucoside with β -1-6 linkages, by a random mechanism. Glucose and gentiobiose were the end products of this reaction. The enzyme had no activity on cellulose or on crown gall polysaccharide, the latter having mainly β -1-2 glucosidic linkages. The findings confirm the presence of β -1-6 and β -1-3 bonds in the glucan layer of baker's yeast. The β -1,6 glucanase and β -1,3 glucanase have been completely separated from each other on DEAE-cellulose columns. The action of the separated enzymes on walls of different yeasts is being studied.

XV. Institute of Food Technology & Microbiology, College of Horticulture & Viticulture, Budapest, XI, Ménési ut 45. Communicated by Dr. K. Vas.

Two years ago I left the Institute of Research in Canning and Refrigeration to become Professor of Food Technology and Microbiology in the College of Horticulture and Viticulture, Budapest. On several problems of microbiological interest, we work together with my former institute, now reorganized into the "Central Food Research Institute", Budapest.

Our work comprises, among other things, some aspects of yeast physiology and biochemistry.

A study of the gas production of yeasts aims at elucidating some problems of the obscure process of gas development in yeast cultures. The quantitative aspects of the phenomena of the beginning of gas production are under study. It has been established that the cell count of cultures of baker's yeast just beginning to show signs of fermentation is fairly constant, whatever the initial cell count of the inoculated medium may be. A linear correlation could be shown to exist between the logarithm of the initial count (N_0) and the time of appearance of the first visible bubble (T , hours): $T = 31.55 - 4.18 \log N_0$ in the range $N_0 = 10^3$ to $10^{6.5}$. Cell counts measured at the time T (N_T) show a very slight increase (between $10^{6.5}$ and 10^7) as the initial count is raised in the above range. In tubes with N_0 values greater than 10^7 , gas production occurs without further growth. If the extent of multiplication during T is expressed by the ratio N_T/N_0 , the logarithm (to base 2) of this value shows a linear correlation with T : $T = 2.42 + 1.41 \log (N_T/N_0)$, demonstrating the fact that growth occurs at an even rate at all initial counts (below $10^{6.5}$), up to the first appearance of gas production, the generation time being 1.41 hr. Kovacs-Proszt, Mrs. G. & Vas, K: Study of the gas production of Sacch. cerevisiae, paper presented before the III Congress of the Society of Hungarian Microbiologists, Budapest, October 4, 1961.

In a second project, investigations were made into the thermal decomposition of yeasts (Schizosacch. pombe, Sacch. cerevisiae, Sacch. rouxii, Hansenula anomala, Kloeckera apiculata).

Thermal decomposition of a clean, thoroughly washed, dry cell mass, mixed with Al_2O_3 , was examined in the temperature range between 20 and $1000^\circ C$, with the aid of the so-called derivatograph of Erdey, Paulik and Paulik (Nature, 174, 885, 1954). The loss in weight, the rate of weight loss, and the heats of reaction of the decomposition processes caused by heat treatment were measured on the cell mass placed in a space heated at a predetermined rate (approximately $10^\circ C$ per minute). All the above changes were automatically recorded as a function of time.

Apparently, considerable differences exist between the behavior of each microorganism. We presume that these differences may be used as characteristics in the identification of the microorganisms, as "finger prints" in taxonomy.

The method seems especially suitable for the examination of the insoluble components and structures of the cell. Studies of thermal decomposition of the cell promise to be a means suitable for the chemical characterization of the insoluble matter of the cell, as well as of its major soluble components.

A more detailed account of these results has been published in Hungarian, in the Communications of the Central Food Research Institute (Vas, K. & Proszt, G.: Investigations into the Thermal Decomposition of Microorganisms, KEKI-Közl., 1961. No. I., p. 1-5).

In a third field of research, we are interested in the effect of ionizing radiations on yeast cells. Some marked effects of X-rays and cathode rays on 20% suspensions of Sacch. cerevisiae cells were examined, 1) by measuring the leakage of free amino acids from the cells into the surrounding aqueous phase, and 2) by measuring changes in dehydrogenase activity of the suspension by the triphenyltetrazolium chloride method. We suggested

(Vas, K. & Farkas, J.: KEKI-Közl., 1960, No. 1, p. 1-5) that the primary effect of irradiation is the destruction of the cells (the capacity of growing is lost) causing disorganization of the cell and a decrease in dehydrogenase activity. With increasing dosages the cytoplasmic membrane is damaged, its permeability increases, the leakage of amino acid and the penetration of TTC is accelerated. At extremely high doses the enzyme-inactivating effect of irradiation becomes apparent.

Work on microbiological stabilization of fruit juices showed that film-forming yeasts (e.g. Hansenula) are less resistant to ionizing radiation than purely fermenting species (Sacch. cerevisiae). It has been observed that in contrast to the thick and rough appearance of the film formed on original fruit juice, those grown on juices not sufficiently treated for complete inhibition are very thin and smooth structures. The cause of this phenomenon remains to be elucidated (Vas, K. & Farkas, J.: Annales de l'Institut Pasteur de Lille, 11, 209, 1960).

Our recent investigations showed that combination of irradiation with mild heat treatment results in an enhanced antimicrobial effect. It was interesting to note that irradiation prior to heat treatment was much more efficient than heat treatment before irradiation (Farkas, J., Vas, K., & Kiss, I.: Paper presented before the III Congress of the Society of Hungarian Microbiologists, Budapest, October 4, 1961).

XVI. Brief News Items:

1. The Editor announces with deep regret the death of Professor S. Hestrin in February, 1962. Professor Hestrin was a member of the faculty at the Hebrew University of Jerusalem, Department of Biological Chemistry. His untimely death constitutes a severe loss to the scientific community and to his friends.

2. Since the last publication of the Yeast News Letter, the following articles have been published or have been accepted for publication:

Lindegren, C. C., Bang, Y. N. and Hirano, T. Progress report on the zymophage. Proc. N. Y. Academy of Science, 24: 540-566 (1962).

Lindegren, C. C. A proposal concerning the origin of the endoplasmic reticulum. Nature 195: 1225 (1962).

Extra copies of "Yeast Genetics 1962" are available for distribution to anyone interested in receiving them. Dr. Lindegren has signed a contract with Prentiss-Hall to write a book on "Yeast Genetics" for the Foundation of Modern Biology Series.

Carl C. Lindegren, Director
Biological Research Laboratory
Carbondale, Illinois

3. The following article has been published recently:

"Les levures à spores réniformes", J. Boidin, F. Abadie, J. L. Jacob et M. C. Pignal, Bulletin Trimestriel de la Société Mycologique de France, Tome 78 (2), 155-203, 1962.

I would like to exchange this article for others of a similar nature with readers of the Yeast News Letter.

Professor J. Boidin
Laboratoire de Microbiologie et Mycologie
16, Quai Claude-Bernard
Lyon 7e, France

4. The following articles have been published or will be published soon:

Published: Lycette, R., and L. R. Hedrick (1962). Action of Deflocculating Agents on Saccharomyces cerevisiae Class III Brewer's Yeast. Appl. Microbiol. 10: 428-430.

In Press: Lycette, R., and L. R. Hedrick (1963) Adsorption and Fluorescence of Fat Soluble Fluorescent Dyes Upon Class I and Class III Saccharomyces cerevisiae. J. Bacteriol. 1963.

In Preparation: Nero, L., Mae Goodwin-Tarver and L. R. Hedrick. Growth of Acanthamoeba castellanii with the Yeast Torulopsis famata.

Dr. Leslie R. Hedrick
Illinois Institute of Technology
Department of Biology
Chicago 16, Illinois

5. 1) The Instituto de Micologia, Univ. do Recife (IMUR), is carrying out a vast program of yeast survey of the soils of the Amazon basin. The soil samples are obtained from soil profiles opened inside of the virgin and secondary forests at uniform levels: 5, 15, 30, 40 and 60 cm deep.

2) J. L. Bezerra from IMUR spent 6 months working with Prof. J. A. von Arx in Holland and came back to Recife last October.

3) Dr. S. K. Shome after two years of residence at IMUR went back last July to India, his country of birth.

4) A. C. Batista, S. K. Shome and F. M. Santos isolated from soil for the first time the fungus Paracoccidioides brasiliensis, proving also that the samples were pathogenic for animals (IMUR, Publ. No. 373, 1962).

Prof. A. Chaves Batista
Universidade do Recife
Instituto de Micologia
Recife, Pernambuco, Brazil

6. The following papers have been published by members of our Institute:

Verona, O. and A. Rambelli. Intorno ad un ceppo di Candida reukaufii (Grüss) Diddens et Lodder isolato da fiori di Eucalitto. Atti Istituto Botanico Lab. Critt. Università Pavia, Serie 5, Vol. XIX, pag. 77-84 (1961).

Verona, O. and A. Rambelli. Notizie, ricerche e considerazioni relative Candida pulcherrima ed altri lieviti ad analoga fisionomia. Annali Facolta di Agraria, Vol. XXII, 1961, pg. 92-121.

Aquarone, E. Qualche ricerca su di alcuni lieviti osmofili. Annali Facolta di Agraria, Vol. XXII, 1961, pg. 195-214.

Verona, O. and G. Picci. Sul potere azoto-fissatore di Candida pulcherrima. Annali Facolta di Agraria, Vol. XXII, 1961, pg. 233-234.

Verona, O. and A. Rambelli. About two isolates labelled Taphrina farlowii Sad. and Taphrina californica Mix. Phytopathologische Zeitschrift, Band 44, Heft 3 (1962), S. 269-272.

Prof. Onorato Verona
Istituto di Patologia Vegetale
e Microbiologia Agraria dell' Universita
Via S. Michele, 6
Pisa, Italy

7. I have a graduate student working on a yeast which buds on a broad base. It looks as though it may be a Saccharomyces. We would very much appreciate receiving cultures of similar organisms that people may have in their collections, and we would like to correspond with people who may be studying them.

Dr. Alfred F. Borg
Department of Bacteriology
Kansas State University
Manhattan, Kansas

The Editor expresses his warmest wishes to the readers of the Yeast News Letter for a Happy and Prosperous New Year.

H. J. Phaff