YEAST

A News Letter for Persons Interested in Yeast

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The Editor takes pleasure in thanking all those who have contributed to this issue. Without this gratifying support the News Letter cannot fulfill its purpose. The Editors would like to invite others to send in contributions for future issues. It is planned to publish the next issue of the News Letter in November 1958. It would be appreciated if anyone would notify the Editor of additional people in our field who would like to receive the Yeast News Letter.

I. South African Council for Scientific and Industrial Research, National Chemical Research Laboratory, Wine Industry Research Group, Stellenbosch, C.P. Communicated by Dr. J. P. van der Walt.

Brettanomyces in South African Table Wines.

The South African Wine-industry has, like all other wine producing localities, long been acquainted with the sporadic occurrence of yeast turbidities in table wines. However, during the last decade it is reported that there has been a marked increase of this type of spoilage for reasons not yet quite apparent. In a survey of the causative organisms, the remarkable fact came to light, that apart from the ascosporogenous yeasts commonly associated with turbidity, no less than 50% of such affected wines were found to harbour Brettanomyces species. Moreover, this infection occurs not only in dry, white wines but also in semi-sweet, white table wines, dry and medium sparkling wines, dry sherries and also in dry, red wines. The infection occurs in both quality and cheaper wines alike.

An incidence of 50% is rather unusual if it is borne in mind that Dr. Simone Domercq only reports an incidence of about 2% for the wines of the Gironde.

New Yeasts:

During the study of yeasts associated with the larval feed of the South African bumble bee, an interesting and hitherto unknown ascosporogenous yeast species was encountered.

The strains form small round to short-oval vegetative cells which are predominantly haploid. In liquid media a transient pellicle is formed. Pseuodmycelium formation is absent.

A heterogamous or isogamous conjugation precedes ascus-formation. The ascospores, 1-4 per ascus and brownish in colour, are remarkable in that they are oblate ellipsoidal - a shape not previously encountered in the yeast domain. The ascospores stain an intense yellow with iodine.

The assimilations of both nitrate and nitrite are absent. The species is strongly fermentative, fermenting glucose, saccharose, maltose and raffinose 1/3.

The classification of the species is at present not possible and is awaiting more fundamental investigations regarding its phylogeny.

- II. Department of Fermentation Technology, College of Agriculture, University of Ankara, Ankara, Turkey. Communicated by Dr. Huseyin Sahinkaya.
- I. College of Agriculture, University of Ankara (publication No. 114, 78 pages, 38 illus.) "Experiments upon the flora of microorganisms and especially the wine yeasts in the vineyards at the province of Ankara". Thirty-one pure yeast strains were isolated from musts that had been obtained from various grapes in the area. Some of the morphological, physiological and technological characteristics of these yeasts were determined. All results were compared and the superior strains were chosen. These strains with superior qualities (Kaleckik II, Dikmen, Hasandede) are strains of Sacch. Cerevisiae (they were identified in Dr. L. J. Wickerham's Laboratory).

They are now used widely in the wine industry of Turkey.

II. "Influence of metal ions on growth and metabolism of yeast" This research was carried out in the Dept. of Biochemistry and Nutrition, College of Agriculture, University of Nebraska.

Five Turkish and five American strains of yeast and eleven metal ions (Li, Cu, Ce, Cd, Mn, Cr, Ni, Fe, Zn, Mo, V) were used.

First, the effect of single metal ions and then their combined effects (two or three together) were examined. Very interesting results were found. For example, Mn ions counteract the toxic effect of Cu and the toxic effect of Cu is increased by adding Mo ion. When Cu and Cd are present together, their toxic effect is increased. The toxicity is counteracted by adding Mn to the medium.

I left Tarkey on October 1955 for two years doing research at the University of Nebraska, Department of Biochemistry and Nutrition. I worked there until November 1957. After my return I continued my work on the role of trace elements on growth and metabolism of yeasts. The brief description of the method and results will be given in "Nature".

III. Section of Microbiology, Institute for Research in Canning, Meat Packing and Refrigeration, Budapest, II., Herman O. -ut 15, Hungary. Communicated by Dr. K. Vas.

Effects of Deficiency in Metal Ions on Cell Form and Kinetics of Growth of Two Species of Saccharomyces.

Abstract of a paper prepared for publication by G. Gantner & K. Vas.

Changes in form and growth occurring in media treated on cation exchangers have been studied with two yeasts, the wine yeast Sacch. cerevisiae var. ellipsoideus, strain "Tokaj 22" (obtained from the yeast collection of the Hungarian Ampelological Research Institute, Budapest) and an osmophilic yeast Saccharomyces rouxii, Boutroux isolated from tomato puree in our laboratory (Vas & Proszt, Elelmezesi Ipar, 9, 1955, 41). In contrast to similar experiments made earlier with apple and grape juices (Vas, Acta Chim. Hung., 3, 1953, 339), the present investigations were performed with tomato juice. The clear serum, obtained after elimination of the fibers, was treated with Dowex 50 cation exchanger. This "treated" juice was then remixed with "untreated" serum in various proportions and sterilized by heating in steam.

It was found that, as regards changes in form, the two species behaved differently. After calculating the frequency distributions of cell volumes, statistical analysis showed that in "treated" juices the mean of cell volumes of Sacch. cerevisiae decreased significantly. The ratio of their longer and shorter axes increased causing the cells to become slightly more slender in general. It is worth mentioning that, as stated in an earlier report (Vas & Proszt, 1956 in press), here too the frequency distribution of cell volumes proved to be log-normal in nature. —The volume and form of cells of Sach. rouxii changed much less. No statistically significant differences could be observed. Thus, the latter species seemed to be more resistant to changes in form than the wine yeast studied.

Kinetic measurements of growth, effected turbidimetrically (Vas, Acta Microbiol. Hung., 2, 1955, 215 & 235), showed the osmophilic yeast to be the slower-growing one. In juices treated with the cation exchanger, practically no growth occurred. Growth could not be observed in mixtures containing less than 15% v/v original (not treated) juice. With slightly higher ratios of untreated juice, rapid increase in growth occurred. Total growth (Monod, Ann. Rev. Microbiol., 3, 1949, 371) in a mixture containing 20% v/v of untreated juice almost reached the level experienced in a 100% untreated juice sample, while the rate of growth increased only after admixture of more than some 30 or 40% v/v of original juice to the treated one.

IV. Soil Bureau Experimental Station, Department of Scientific and Industrial Research, Eastern Hutt Road, Lower Hutt, New Zealand. Communicated by Dr. Margaret di Menna.

Work on soil yeasts is continuing. Results from this last season have confirmed previous findings upon yeasts on the leaves of pasture grasses. During winter and spring the phyllosphere supports moderate yeast populations, largely of colourless species and mainly of Cryptococcus laurentii. For what seems to be a relatively short period, of the order of a few weeks, in the late summer or early autumn, there is a great increase in numbers and the population is dominated by species of Rhodotorula. Three strains of Candida albicans were recovered from one grass sample taken in the summer. The pasture from which the grass was collected was grazed by sheep, and other mammals known to carry C. albicans. One of the strains has been sent to Professor M. Silva of the College of Physicians and Surgeons, Columbia University.

Yeasts from some samples sent from Antarctica have been examined. Moss, which was received six days after collection, supported a yeast flora of 75,000 per gram. Cryptococcus laurentii was the commonest isolate. Counts on plates incubated at 4°C were the same as on those incubated at room temperature.

Some samples of Antarctic silt in which algae were growing took rather longer to reach the laboratory but had been kept frozen in transit. No yeasts appeared on cultures incubated at room temperature, but some appeared on plates held at 4°C. These yeasts must be truly psychrophilic, for exposure to a temperature of 20°C for two days seems to kill them. Their identification is going to be protracted as all cultures are having to be incubated in a refrigerator for a much longer time than usual.

V. <u>Institut de Recherches pour les Industries du Cuir. 181 Avenue Jean-</u> <u>Jaures, Lyon, France.</u> <u>Communicated by Dr. J. Boidin.</u>

Deux Curieuses Levures Isolees D'Extraits Tannants D'Origine Vegetale: Pachysolen (nov. gen.) tannophilus nov. sp. et P. Pelliculatus nov. sp.

by Jacques BOIDIN and Jose-Maria ADZET
Appeared in "Bulletin de la Societé Mycologique de France, 73 (4), 331-342,
1958. 5 figures and 2 photos, 1957.

Five strains have been isolated from concentrated tanning liquor, which show very remarkable peculiarities: the Mother ascus cell sends out

a thick walled tube, while its own normal wall also becomes thicker. Only the extremity of the tube has a thin wall and contains all the cytoplasm and the nucleus. At the end of the tube, which varies in length from 3 to more than 100 microns, a swelling develops which soon contains 4 subhemispherical, hyaline ascospores. P. tannophilus has the following characteristics:

Wall wall

glucose fermented slowly; glucose and galactose assimilated; nitrate assimilated; ethanol -; arbutin -.

P. pelliculatus differs only slightly from the first species by forming a pseudomycelium and a pellicle on malt extract. It, practically, has no fermentative power.

The proposed introduction of this new genus, characterized by significant morphological features and its sexual reproduction, in the subfamily Lipomycetoideae Lodder et Kreger van Rij, necessitates a redefinition of this subfamily.

Strains No. 145 (P. tannophilus) and No. 164 (P. pelliculatus) have been deposited at the CBS culture collection at Delft (The Netherlands).

VI. University of Kentucky, Department of Zoology, Lexington, Kentucky.

Communicated by Dr. John M. Carpenter.

Studies on the Ecology of Drosophila

Over 320 strains of yeast have been isolated from the crops of the five species of <u>Drosophila</u> undergoing analysis in this phase of the problem. The identification of these strains to genera and species has been underway for approximately ten months. This phase of the work, as anticipated, has been the most difficult to initiate, it being necessary to learn, and in some cases develop, new techniques. The help of Dr. L. J. Wickerham has been of great aid in this part of the work.

At the present time, nine yeasts from <u>Drosophila</u> crops have been identified to genera. These genera are <u>Candida</u>, <u>Debaryonyces</u>, <u>Hansenula</u>, <u>Hanseniaspora</u>, <u>Kloeckera</u>, <u>Pichia</u>, <u>Rhodotorula</u>, <u>Saccharonyces</u>, and <u>Torulopsis</u>. There are other, but tentative, generic identifications. Five yeasts have been identified to species, those being: <u>Hansenula anomala</u>, <u>Pansenula subpelliculosa</u>, <u>Kloeckera apiculata</u>, <u>Pichia fermentans</u>, and <u>Pichia membranae-faciens</u>. These identifications to species, which are difficult to make, are tentative however and will be checked with Dr. Wickerham before definite determinations are published.

There is some indication that certain fly species prefer certain yeasts, e.g., <u>D. affinis</u> seems to harbor more <u>Kloeckera</u> than other genera, <u>robusta more Pichia</u>, etc. More data is needed, however, in this study. There is also beginning to appear in our data some indication of seasonal yeast fluctuations, but again more data is needed.

Papers indicating the results just briefly summarized have been given by members of the research staff at several meetings during the past two years. These papers number eight and have been presented at two state academy meetings, two regional meetings and at one national meeting.

One paper is at present being prepared for publication. Three abstracts concerning the research have already been published.

VII. Carlsberg Laboratorium, Copenhagen, Denmark. Communicated by Dr. Catherine Roberts.

During this year work on yeast cytology has been continued, and Mr. A. T. Ganesan has completed an investigation of the cytology of the haplophase and the diplophase in Saccharomyces. Particular attention has also been paid to the nucleolus, both in Lipomyces and in Saccharomyces, as well as to the occurrence of giant cells in these two genera. Dr. van Uden's culture of Saccharomyces italicus var. melibiosi is being employed in a detailed study of the genetics of melibiose fermentation, which is being carried out by Mr. Ganesan and Dr. Wolfgang Haupt from the University of Tübingen. Other guests this year have included Hr. Cand. real. T. A. Pedersen from the Mikrobiologisk Institutt in Vollebekk, Norway, who was here for a month in order to study an as yet unidentified imperfect yeast isolated from forest soil, and Dr. S. Reiersol from Statens Institutt for Folkehelse in Oslo, who acquainted herself with techniques employed in yeast genetics. A publication entitled "Some Observations on Sporulation in Saccharomyces" by A. T. Ganesan, H. Holter, and C. Roberts is now in press.

VIII. Cytogenetics Laboratory, Indian Institute of Science, Bangalore-3. India. Work carried out under the guidance of Dr. M. K. Subramanian. Reported by T. R. Thyagarajan.

- 1. Royan, S., & Subramaniam, M. K.
- 2. Royan, 5.
- 3.
- 4.
- 5. "
- 6. " & Bhattacharya, D.L.
- 7. Aswatha Narayana, N. V.
- 8.

"The Nucleus in the Living Yeast". Proc. Ind. Acad. Sci. 1956, 43B, 228. "Variations in the Structure of the Nucleus in Living Yeast". Proc. Ind. Acad. Sci. 1956, 44B, 47. "Structures Revealed by Dark Ground Illumination in Living Yeast". Proc. Ind. Acad. Sci. 1956, 44B, 171. "The Relation between the Nucleus and the Vacuole in Yeast". Curr. Sci. 19**56, <u>25</u>, 3**97. "Phenomena preceding Sporulation in Schizosaccharomyces octosporus". Proc. Ind. Acad. Sci. 1956, 44B, 311. "Some Cytoplasmic Detailes of Yeast revealed by Electron Microscopy". Curr. Sci. 1956, <u>25</u>, 155. "The Tonoplast in Yeast". Curr. Sci. 1956, 25, 88. "The Vacuole in Yeast". Proc. Ind. Acad. Sci. 1956, 43B, 314.

9. Thyagarajan, T. R.

10. Subramanian, M.K.

11. " & "

12. Royan, S.

13. Thyagarajan, T. R.

14. "

15. Aswatha Narayana, N. V.

"Some Observations on Living Cells of Saccharomyces carlsbergensis from Twenty Four Hour Agar slants with Phase Contrast and Dark Ground Illumination". Proc. Ind. Acad. Sci. 1956, 44B, 278.

"The Structure of the Nucleus in Saccharomyces carlsbergensis". Naturwiss. 1957, 44, 68.
"The Nucleus of the Living Zygote of Saccharomyces carlsbergensis". Proc.

Ind. Acad. Sci. 1957, 45B, 187.

"Evaluation of Staining Procedures following Fixation of Yeast in Iodine-formol-acetic solution". Proc. Ind. Acad. Sci. 1958, 47B, 31.

"The Reaction of the Vacuole and the Nucleus of Living Yeast to Neutral Red". Curr. Sci. 1958, 27, 28.

"Spore Germination and Zygote Formation in Saccharomyces carlsbergensis". Proc. Ind. Acad. Sci. 1958, 47B, 124.

"The Behaviour of the Vacuole on Stimulation of Yeast Cells with Fresh Media".

Proc. Ind. Acad. Sci. 1958, 47B, 225.

IX. Department of Biology, Western Reserve University, Cleveland 6, Ohio. Communicated by Thomas D. Brock.

Research in progress.

- 1. Studies on the mechanism of agglutination of mating types of Hansenula wingei have been carried out and are still in progress. A paper on preliminary work will appear in the June issue of the Journal of Bacteriology. Current evidence indicates that agglutination of mating types when mixed is due to attraction between a specific protein component of one strain (#21) with a specific complementary non-protein, probably polysaccharide component of the other strain (#5). Thus the agglutination is analogous to an antibody-antigen reaction. Chemical studies on the yeast cell surface are now underway to characterize these components in detail.
- 2. Inhibition of yeasts by 2,3,5-triphenyl tetrazolium chloride (TTC). TTC has been used by many people as an oxidation-reduction indicator and has been often incorporated into agar, since colonies which reduce it will become red. This phenomenon has been used to screen for fermentation mutants. So far as I know, there has been no report of TTC inhibiting yeast growth. I found quite accidentally that TTC was highly inhibitory to certain yeasts, but had no effect on others. The data in the accompanying table show results of tests on agar plates incorporating TTC.

Organism	Minimum Inhibitory Concentration, pg/ml	Organisa	Minimum Inhibitory Concentration ug/ml
Sacch. priorianus	8	Hansenula subpelliculosa	4
Sacch. cerevisiae	64	H. beckii	8
Sacch. carlsbergensis	125	H. wingei	16-64
Sacch. lactis	125	H. canadensis	250
Sacch. fragilis	250	H. capsulata	500
Endomyces magnusii	250	H. saturnus	500
Schizosach, octosporus	250	H. californica	1000
Rhodotorula glutinis	500	H. anomala	1000
Pichia membranaefaciens Candida utilis	500 1000	H. ciferri	1000

The wide range of sensitivities within the genera Saccharomyces and Hansenula is quite interesting, and as yet I have no explanation to offer. There is absolutely no correlation between ability to reduce TTC and sensitivity. Both H. anomala and H. subpelliculosa are powerful reducers of the compound.

A brief note on the effect of TTC on yeast growth will appear in a forthcoming issue of Die Naturwissenschaften.

- 3. A graduate student in my laboratory, Dale Hunt, is working on the mode of action of the antibiotic Actidione on yeasts. Since the genus Hansenula is highly sensitive to inhibition by this compound, he is working with <u>H. wingei</u>.
- 4. Anyone passing through Cleveland is cordially invited to stop and visit us. Western Reserve University is in the cultural heart of Cleveland, and there are many worthwhile things to see.
- X. Universidad De La Republica, Facultad de Hunanidades y Ciencias, Cerrito 73, Montevideo, Uruguay. Communicated by Dr. Noemi Garcia-Zorron.

This is a surmary of a paper entitled "Inheritance of the Ability to Hydrolyze Cellobiose and Melezitose in <u>Hansenula subpelliculosa</u>". This paper was presented at the third South American Botanical Congress. Three crosses were made between strains which are differentiated by their behavior towards cellobiose and melezitose. The hybrids obtained were studied, as well as their segregations. As a first result of the three crosses realized, we conclude the existence of genetic factors responsible for the ability of the several strains to hydrolyze melezitose and cellobiose. In both cases the abilities to hydrolyze these carbohydrates appear as dominant factors. To explain the three segregations studied, the hypothesis is proposed that there exist in <u>Hansenula subpelliculosa</u> two polymeric genes responsible for the hydrolysis of cellobiose, C₁ and C₂ and two polymeric genes for the hydrolysis of melezitose Me₁ and Me₂.

XI. <u>Departamento de Microbiologia</u>, <u>Instituto Botanico</u>, <u>University of Lisbon</u>, <u>Portugal</u>. <u>Communicated by Dr. N. van Uden</u>.

Since our last communication to the News Letter (volume V, number 2)

work on yeasts associated with warm blooded animals has continued and the following studies have been completed:

1. N. van Uden & L. Assis-Lopes. Fermentation of raffinose in the absence of invertase by <u>Saccharomyces italicus</u> Castelli var. <u>melibiosi</u> nov. var.. Portug. Acta Biol., Serie A, 4, 323, 1957.

Nine strains of this yeast were isolated from human faeces and one from the caecal contents of a horse. It seems to be the only known wild yeast; which ferment melibiose and the galactose part of raffinose without fermenting sucrose.

NOTE. Kreger-van Rij found however that some of our strains may ferment sucrose, the hydrolysis probably being done by a maltase. Apparently the yeast extract fermentation medium used in Delft brings out this effect more easily than Wickerham's fermentation medium used in our laboratory.

2. N. van Uden & L. do Carmo Sousa. <u>Candida slooffii</u> nov. sp. a thermophilic and vitamin deficient yeast from the equine intestinal tract. Portug. Acta Biol., Serie A, <u>5</u>, 7, 1957.

This yeast was isolated from the caecal contents of six equines out of a group of 252. Its fastidiousness (all strains are deficient for biotin pantothenate, inositol, niacin, pyridoxin and thiamin when tested at 37°) and particularly its narrow temperature range (growth occurs only between 28° and 44°) are characteristic for microorganisms adapted to warm blooded hosts. Cells are big and a well developed pseudomycelium with branched verticils of blastospores is formed. Only glucose is fermented and assimilated.

NOTE. Meanwhile we isolated Candida slooffii from 121 out of a group of 250 unselected pigs obtained from various parts of Portugal. This unusual high incidence points to pigs as the natural host for Candida slooffii; it is similar to the situation which is found in mice and rats with regard to Torulopsis pintolopesii and in rabbits with regard to Saccharomycopsis guttulata.

3. N. van Uden & L. do Carmo Sousa, Yeasts from the equine caecum (to be published).

From the cacca of 252 adult equines 146 yeast strains were isolated, distributed as shown (in brackets) among the following species: Saccharomyces carlsbergensis (5), S. cerevisiae (8), S. italicus var. melibiosi (1), S. tellustris (2), Pichia membranaefaciens (1), P. fermentans (1), P. farinosa (1), Hansenula anomala (1), Debaryomyces kloeckeri (1), Torulopsis glabrata (3), T. pintolopesii (1), Cryptococcus neoformans (1), Candida albicans (11), C. krusei (21), C. tropicalis (11), C. parapsilosis (9), C. slooffii (6), C. guilliermondii (4), C. bovina (1), C. utilis (1), C. brumptii (1), Trichosporon cutaneum (55).

The incidence of Candida albicans and the Glabrata-group was very low, indicating a low degree or absence of host suitability for these species. The latter are possibly passers-by coming from more suitable hosts (man, small rodents, pigeons, pigs). Equines seem to constitute the most suitable host for Trichosporon cutaneum so far reported. The isolation of a strain of Cryptococcus neoformans from a healthy animal suggests that equines some times are vectors for this pathogen.

Temperature relations, vitamin requirements and assimilative properties of type strains, equine strains and other strains (total number in brackets) of the following species were determined: Saccharomyces tellustris (3), Torulopsis glabrata (6), T. pintolopesii (10), Candida bovina (6), C. slooffii (6), Cryptococcus neoformans (23) and Trichosporon cutaneum (12).

4. N. van Uden, L. do Carmo Sousa & M. Farinha, Yeasts from the caprine and the ovine caecum. (To be published)

From the caeca of 250 adult goats 16 yeast strains were isolated distributed as shown (in brackets) among the following species: Saccharomyces cerevisiae (4), S. microellipsodes (1), Nematospora coryli (1), Torulopsis glabrata (2), Candida albicans (2), C. krusei (4), C. tropicalis (2).

From the caeca of 503 adult sheep 35 strains were isolated:

<u>Saccharomyces fragilis</u> (1), <u>Candida albicans</u> (21), <u>C. krusei</u> (6), <u>C. tropicalis</u>
(3), <u>C. parapsilosis</u> (3), <u>Rhodotorula mucilaginosa</u> (1).

The suitability of goats and sheep to constitute hosts for yeasts of any species seems very reduced.

The sixth International Congress on Tropical Medicine and Malaria will meet in Lisbon from September 5 to 13, 1958. There is a mycological section. Simultaneously the International Society for Human and Animal Mycology will meet in Lisbon. More than fifty mycologists including some well known yeast people have so far announced their coming. Anybody interested in these meetings should write without delay to: Prof. Dr. R. Vanbreuseghem, General Secretary of the International Society for Human and Animal Mycology, Institut de Médicine Tropicale, Nationale Straat 155, Antwerp, Belgium.

XII. Northern Utilization Research and Development Division, U. S. D. A., Peoria, Illinois. Communicated by Dr. L. J. Wickerham.

We have been receiving from clinical laboratories over the past few years an increasing number of cultures of <u>Torulopsis glabrata</u>. We published in J.A.M.A. a short paper on identification of this species. Technicians can readily identify it because it produces no hyphae, it assimilates glucose and trehalose, but not maltose or sucrose. It also produces a gaseous fermentation of glucose and trehalose.

Publications:

"Apparent increase in frequency of infections involving <u>Torulopsis glabrata</u>" by L. J. Wickerham. Jour. Am. Med. Assoc. <u>165</u>, 47 (1957).

"Presence of nitrite-assimilating species of <u>Debaryomyces</u> in lunch meats" by L. J. Wickerham. Jour. Bact. 74, 832 (1957).

- XIII. Columbia University, College of Physicians and Surgeons, Department of Dermatology, 630 West 168th Street, New York 32, N. Y. Communicated by Dr. Margarita Silva.
 - 1. We are sorry to report that Prof. Rhoda W. Benham, former contributor to the Yeast News Letter passed away in January 1957 after a

long illness that had forced her premature retirement from her position as head of the Mycology Laboratory and Associate Professor in the Department of Dermatology, College of Physicians and Surgeons, Columbia University. Her many contributions to our knowledge of yeasts of medical interest as well as her graduate course in medical mycology, the first one organized in this country, are well known by many readers of this News Letter.

- 2. Upon Dr. Benham's retirement in 1956, Dr. Margarita Silva, formerly Research Associate, was promoted to the rank of Assistant Professor and asked to assume Dr. Benham's duties as head of the Mycology Laboratory.
- 3. Recent publications of interest to enzymologists:
 (a) Benham, R. W. Species of Candida most frequently isolated from man: methods and criteria for their identification. J. Chron. Dis., 5: 460-472, 1957.
- (b) Pollack, J. D. and Benham, R. W. The chlamydospores of Candida albicans: comparison of three media for their induction. J. Lab. & Clin. Med., 50: 313-317, 1957.
- (c) Dobias, B. Moniliasis in pediatrics. A.M.A. J. Dis. Children, 94: 234-251, 1957.
- 4. Current research with various members of our group include a search for human carriers of <u>Cryptococcus neoformans</u>; isolation and study of the physiological effect on laboratory animals of a toxic extract from the cells of <u>Candida albicans</u>.
- 5. Among the yeasts routinely isolated from clinical specimens, Candida parapsilosis and Torulopsis glabrata show an apparent increase in incidence in this laboratory (cf. Section XII).
- XIV. Institut fur Garungsgewerbe, Mikrobiologische Abteilung, Seestr. 13, Berlin N 65, (West). Communicated by Dr. Siegfried Windisch.

In 1957 numerous beer yeasts were investigated for the occurrence of cytoplasmic respiration-less mutants. Under brewery conditions such mutations occurred in small numbers. Laboratory experiments designed to compare normal yeasts and respiration-less strains have not lead to clear cut results. Nevertheless, cases have been observed in which considerable numbers of mutants have been obtained. In this way the mutations due to various causes could be defined more exactly. (Vg.: H. Gutz, Naturwiss. 44, 545, 1957 and S. Windisch, Wissenschaftliche Beilage der Brauerei 11, 3, 1958). These investigations are being continued.

On the basis of investigations on the acid formation of film forming yeasts the retention of the variety <u>Candida mycoderna</u> var. <u>lafarii</u> is rejected, because it has been shown that <u>Candida mycoderna</u> consists of a range of strains which gradually vary from weak to strong acetic acid formation. (S. Windisch, Branntweinwirtschaft <u>80</u>, 21, 1958).

Experiments on the causes of pellicle formation by film yeasts have shown a close relationship between cell multiplication and film formation. Quantitative experiments with a known number of cells used as inoculum under identical conditions have shown that the film yeasts multiply

under submerged conditions up to a certain limited cell density. They then form a pellicle and finally show again a strong growth. With surface active substances, such as Tween 80, it is possible to suppress film formation without influencing respiratory metabolism. (S. Windisch and C. Emeis, Wissenschaftliche Beilage der Brauerei 11, 1958 in press). The investigations are being continued.

Dr. Siegfried Zaake has completed a dissertation on the subject "The Behavior of Yeast-like Fungi with respect to Tartaric Acid" (in press: Zbl. fur. Bakt. II). 335 strains belonging to 19 genera of yeastlike fungi have been tested for their ability to use tartaric acid as a single carbon source. Only fungi of the genera Rhodotorula, Cryptococcus and Trichosporon are able to utilize this compound. A few other yeasts were stimulated by tartaric acid. The occurrence of Rhodotorula-like forms among strains of Cryptococcus confirms the relationship which apparently exists between these two genera. To the common physiological properties, the ability to assimilate tartaric acid may be added. The formation of carotenoid pigments in Rhodotorula and the formation of the typical starchlike compounds (which are colored by iodine) in Cryptococcus are the two ends of a scale. Along this scale strains of both genera may change gradually. The pH value proved to be an important condition; at high pH values pigment formation is stimulated; at low pH values the formation of starchlike substances is stimulated.

As a continuation of the investigations on the effect of yeasts in the intestinal canal was studied. The relation of the species to the intestinal bacteria was also determined (F. Staib and S. Windisch. Zbl. f. Bakt. I Orig. 166, 302, 1956).

The occurrence of a surface growth on compressed yeast by Endomyces lactis was the subject of an ecological analysis. The luxurious development of the milk fungus on the compressed yeast is the result of a collaboration without competition with the lactic acid bacteria; the milk fungus contributes panthothenic acid and riboflavin in considerable amounts; the lactic acid bacteria contribute lactic acid, which is utilized preferentially by the milk fungus. (S. Windisch and A. M. Herbst, Branntweinwirtschaft 80, 101, 1958).

In October and November of 1957 I visited the United States of America and saw primarily certain fermentation industries and distilleries as well as several institutes. I remember gratefully the very pleasant reception which I experienced everywhere. Dr. Freeman Weiss of the American Type Culture Collection in Washington D. C. has already pointed out in the Yeast News Letter of November 1957 that I am trying to collect cultures of Spermophthora gossypii. Also cultures of other species of the Spermophthoraceae would be extremely welcome, since I would like to study this group of fungi.

XV. <u>Universidade De S. Paulo, Instituto Zimotecnico, Piracicaba, Sao Paulo, Brasil. Communicated by Dr. Rodolpho de Camargo.</u>

The following projects are under study.

1. Geraldo C. Mello Ayres is working on some physiological aspects of two species of <u>Pullularia</u> isolated from sapoti (a typical Brazilian fruit,

very rich in starch), and from coffee cherries. Mr. Ayres is interested in this genus of yeast and his plans are concerned with the determination of the important biochemical aspects of the genus Pullularia.

- 2. Dr. Jayme R. de Almeida and Rodolpho Camargo work is conducted in order to adapt strains of Saccharomyces cerevisiae and S. carlsbergensis to certain quantities of Emulsan AL (a product commonly used in the alcohol and "cachaca" industries in Brazil) which prevents bacterial contamination in fermentation tanks. The yeasts are now able to support as much as 0.1% of Emulsan AL producing a good fermentation.
- 3. R. Camargo several yeasts were isolated from coffee cherries during Dr. Reese Vaughn's visit to São Paulo last year. The yeasts are now classified.
- 4. Miss S. Joly, with Prof. O. Verona from Piza, Italy, isolated yeasts from regional deteriorated flowers. Many yeasts were obtained and they are now under taxonomic studies.
- 5. Miss R. N. Neder just presented her Doctor's thesis. Subject: Isolation and identification of yeasts from fermenting sugar cane juice for the production of "pinga".
- 6. Mr. A. Serzedello is going to try production of food yeast from sugar cane byproducts.

XVI. <u>University of Wisconsin</u>, <u>Department of Bacteriology</u>, <u>Madison</u>, <u>Wisconsin</u>. <u>Communicated by Dr. H. O. Halvorson</u>.

Since the last publication of the Yeast News Letter the following articles have been accepted for publication:

- 1. Cohen, G. N., Halvorson, H. O. and Spiegelman, S. Effects of p-fluorophenylalanine on the growth and physiology of yeast.
- 2. H. O. Halvorson and L. Ellias. The purification and properties of and inducible yeast & -glucosidase.

Our effort since the last news letter has been concentrating on further defining several related inducible systems in yeast. Miss Sara Winderman has succeeded in purifying an \propto -glucosidase 110 fold and Mr. Duerksen purified a β -glucosidase 120 fold over crude extracts. The β -glucosidase has a SH group involved in overall catalysis of substrate and α -glucosidase has both a SH group and an ionizable group pK 6.8 (histidine?) involved in hydrolysis of α -glucosides. The molecular weight of α -glucosidase is 85,000 and of β -glucosidase, 300,000. The specificity of the C atoms 1-4 of glucose are required for affinity for α -glucosidase, while β -glucosidase demands specificity only at C1, C3 and C4. We are currently examining related α -glucosidases.

XVII. <u>University of Illinois, Department of Bacteriology</u>. <u>Communicated by Dr. F. M. Clark</u>.

We are continuing our work in this laboratory on inositol and

yeast growth. Recently Prof. Phaff was kind enough to send me a culture of Torulopsis melibiosum which assimilates inositol as a sole carbon source in an ammonium-inorganic salts medium. We are studying the breakdown of inositol by this yeast with special interest in intermediate and final products produced from inositol. Our work up to the present has been confined to developing a medium in which the yeast will grow best with the greatest utilization of the carbon source being studied. Relatively poor growth was obtained with Wickerham's Medium using (NH₄) 2 SO₄ as the nitrogen source. Only about 40% of the inositol was assimilated in 72 hours. When ammonium sulfate was replaced by NH₄H₂PO₄ and the reaction adjusted to pH 5.5-6.0 much better growth was obtained with a utilization of approximately 70% of the inositol supplied. We believe this effect is the result of pH since in the ammonium sulfate medium the final pH was much lower than that of the ammonium phosphate medium.

Bacteriologist meeting in Chicago on "Production of Acarotene by species in the genus Rhodotorula". This represented work he did with this group of yeasts for his Doctor's thesis. It is anticipated that this work will be published. Dr. Deufel is now employed at the Eli Lilly Research Laboratories in Indianapolis, Indiana.

XVIII. Department of Food Technology, University of California at Davis. Communicated by Dr. H. J. Phaff.

- 1. Mr. Harry Snyder is engaged in research on an inulin hydrolyzing enzyme produced by Saccharomyces fragilis. It has been found that the enzyme is produced extracellularly and can completely hydrolyze inulin by splitting off fructose units from the end of the chain. The enzyme is inducible and some interesting problems have arisen concerning the conditions under which induction will occur. Also the relationship between β fructosidase of baker's yeast and the inulin hydrolyzing enzyme is being investigated.
- 2. Mr. H. Tanaka, visiting student from Japan, is completing his M.S. degree and has worked on the yeasts and molds which cause spoilage of fresh and dried prunes. The effect of osmotic pressure and relative humidity on the growth of selected spoilage organisms has been determined.
- 3. Mr. J. D. Ferreira from the "Estacao Agronomica Nacional, Quinta do Marquez, <u>Oeiras</u>, <u>Portugal</u>, is working on the life cycle of a new species of <u>Schwanniomyces</u>. Spore germination, nuclear behavior, ploidy and similar aspects are being studied. Mr. Ferreira is in the United States on a Fellowship of the Calouste Gulbenkian Foundation.
- 4. The following two papers have been accepted for publication in "Antonie van Leeuwenhoek".

"On the cell wall composition of the apiculate yeasts" by M. W. Miller and H. J. Phaff.

"On the isolation, ecology and taxonomy of <u>Saccharomycopsis</u> guttulata (Robin) Schionning" by M. Shifrine and H. J. Phaff.

XIX. Brief News Items

It is with deepest regret that I have to inform you that Dr. Charles Edward Skinner passed away on May 10 in Minneapolis. He and Mrs. Skinner were in the Mid-West where he had attended the meetings of the Society of American Bacteriologists. Just before catching the train on May 2 he was stricken with a bulbar stroke, which left him partially paralyzed and became progressively worse.

Dr. Skinner was born in Brainerd, Minn., March 18, 1897. He obtained his B.S. and M.S. degrees at the State College of Washington and his Ph.D. at Rutgers. After obtaining his Ph.D. degree he spent a year in England under an International Education Board Fellowship in Soil Microbiology at Rothamsted. He then taught at the University of Minnesota and in 1948 returned to the State College of Washington at Pullman, where he was Professor of Bacteriology and Public Health and Chairman of the Department until his death. His research included studies on sanitary water analysis, soil algae; vitamin synthesis by microorganisms, microbial proteins as food for animals; amino acid synthesis by microorganisms; microbial tyrosinase; yeastlike fungi; yeasts; pathogenic fungi; coliform bacteria.

B. R. Ray, Associate Dean, Division of Natural Sciences

Dr. Maurice Sussman, now at Northwestern University will join the staff of the Biology Department at Brandeis University as Associate Professor of Biology beginning September, 1958.

> Harold P. Klein, Chairman Department of Biology Brandeis University Waltham 54, Massachusetts

During the British Association meeting at Dublin, Ireland, the Chemistry and Botany Sections held a joint session on September 9, 1957, at which "The Biology of Yeast" was discussed. Professor D. G. Catcheside spoke on "The Genetics of Yeast", Professor E. J. Conway on "Cell Structure and Function in Yeast", Dr. R. Davies on "The Biochemistry of Alcoholic Fermentation" and Mr. R. B. Gilliland on "Yeast Biology in Relation to Fermentation Techniques". A summary of the papers and discussion was published in Nature, 130, 1161-3, 1957.

R. B. Gilliland
Arthur Guinness Ltd.
Chemist's Laboratory
St. James's Gate, Dublin, Ireland

Dr. Carl C. Lindegren was chosen as the eleventh recipient of the Annual Pasteur Award by the Society of Illinois Bacteriologists on February 28 in Chicago. He gave a talk entitled "Recombination in Microorganisms".

On April 30, Dr. Lindegren talked on "Reverse Linkage in Saccharomyces" at the S.A.B. meeting in Chicago.

Dr. Lindegren gave a talk at Detroit on May 7 at the 24th Convention of the American Society of Brewing Chemists entitled "Respiratory Deficiency and the Zymophage; two factors capable of causing degeneration in brewery

yeasts", which will be published in the Proceedings of the American Society of Brewing Chemists.

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

Dr. E. M. Mrak was awarded the Nicholas Appert Award by the Institute of Food Technologists. His address entitled "The World I Lived In" has been published in the journal "Food Technology" XI, 541 (1957). Dr. Mrak was also selected as Faculty Research Lecturer on the Davis Campus of the University of California and spoke on "Some interesting aspects of the ecology of yeasts".

Dr. R. C. Artagaveytia-Allende (Montevideo, Uruguay) writes as follows:

In volume VI No. 1 page 8 (May 1957) of the Yeast News Letter an error should be corrected to avoid misinterpretation of the results. <u>C. pseudotropicalis</u> and <u>T. pintolopesii</u> were isolated from nice and not from rats.

I have left my former position at the Instituto de Higiene. I am now in charge of the Mycology section of the Botany Lab at the Faculdad de Humanidades e Ciencias.

The staff of this laboratory is formed by Botany Professor, Eng. B. Rosengurtt, Miss N. Garcia-Zorron, graduated in sciences, and myself.

The laboratory address is: Laboratorio de Botanica - Faculdad de Humanidades y Ciencias - Cerrito n 13 - Montevideo-Uruguay.

In the beginning of next May I am leaving for the city of Recife to work with Professor A. Chaves Batista on yeasts of medical interest.

I shall work in the Instituts de Micologia da Universidade do Recife where I hope to stay several months.

LETTERS TO THE EDITOR

Dear Sir:

Symbolism in the heterothallic yeasts. Since Blakeslee first discovered mating types in the Mucorales, a number of different symbols have been used to designate the two strains. Blakeslee himself used the symbols \div and \div , but later authors working in other groups of organisms, have seen fit to use different symbols. A quick perusal of the literature has revealed the following symbols: A and B, A and a, a and \heartsuit , mt⁺ and mt⁻, as well as \div and \div . In the yeasts, a and \heartsuit have been used most often. Since all of these symbols refer to essentially the same phenomenon, it would seem desirable to have a standard symbolism of use in all situations.

Certain groundrules for a uniform symbolism can be listed. 1: Mating type symbols should refer to the phenotypes of the isolates, rather

than the genotypes, since it is generally not readily possible to characterize such a complex of physiological factors in terms of single genes. 2: The symbols should be easy to write on any typewriter. 3: The symbols should be easy to speak in oral communication. 4: If possible, priority should be respected.

On all of these counts, it would seem that Blakeslee's original symbols would be the best. I therefore propose that the symbols and - be used to designate isolates (not genes) which can be characterized as mating types. When the genetic aspects of mating type are worked out for any species, the genes can be designated by any desired letters without upsetting this symbolism.

A more extended discussion of this subject with citations to the original literature will be published soon in Mycologia.

Thomas D. Brock
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