YEASTS

A News Letter for Persons Interested in Yeast

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Sufficient news is collected since the last issue to warrent another letter. Furthermore, the editor is leaving on a Sabbatical for six months, so it is about time to get out what has accumulated. I hope that when this one reaches you, you will find it possible to send any new information to Dr. H.J. Phaff, his address is given above, who will carry on. As soon as sufficient information is collected, he will put out another issue.

Cost of operation.

In this connection, the story is the same as it has always been. It is purely on a voluntary basis.

NEWS OF INTEREST

I. New Book to be Fublished by Dr. Ingram in England.

We received an interesting letter from Dr. Ingram located at the Low Temperature Research Station, Downing Street, Cambridge University, Cambridge, England, indicating that he has written a book called "An Introduction to Biology of Yeasts" which is mostly physiological and aims at about the University finals level. Last June it was in the galley proof stage and Dr. Ingram hoped (perhaps optimistically) that it may be published later in the year (1953).

Dr. Ingram also writes that he is hoping to study the assimilation reactions of a number of osmophilic Zygosaccharomyces. In view of the various vitamin requirements noted some time ago by Lochhead and Farrell, Dr. Ingram has the suspicion that Lodder and Kregervan Rij may have been too sweeping in amalgamating these species.

Editors Note: (Dr. Ingram has a real point.)

Or. Ingram has requested that he be placed on the mailing list of the Newsletter, although he is unable to do as much work in the field as he would like to do. It is a pleasure to oblidge in this case and to place his name on the mailing list.

II. Note from the Hebrew University, Hadassan Mccical School.

Dr. Shlomo Hestrin of the Hebrew University, Hadassah Medical School, Department of Microbiological Chemistry, Jerusalem, Israe, writes that his laboratory has temperarily deviated from the straight and narrow, and are now working on levan and cellulose in bacteria with a little effort directed toward algae in fish ponds. At present, they are not doing anything on yeast except for transfer of our cultures, but it is their hope that they may return to yeast sometime in the near future.

III. Studies on yeast at the Illinois Institute of Technology.

Dr. Leslie Hedrick of this institution has sent in the following information:

1. Some studies upon the agglutination of sensitized yeast by the corresponding antibody.

Yaye Furutani, L. E. Klinger, J. Corbett, and L.R. Hedrick. Department of Biology, Illinois Institue of Technology, Chicago 16, Illinois.

Preliminary studies with yeast cells, mostly of the genus Hensenula indicated that yeast cells may be of importance in fundamental

as well as practical studies in the field of immunology. For the most part, living cells are very poor as antigen acceptors. Therefore, the cells must be treated either by heat or by other methods. The treated cells when exposed to certain types of antigens, become sensitized. These when mixed with the appropriate antibody usually give specific agglutinations as revealed either by the microscopic slide, or tube reactions. Some of the antigen-antibody systems that we have tried and that offer some promise are: TB antigen vs. TB antiserum; polysacchoride; type antigens vs. specific antibodies of several bacteria such as Streptococcus pycgenes, D. pneumonia, E. coli, and others.

Some results obtained with different Hansenula species indicate that with the S. pyogenes antigen-antibody system, H. minuta (a haploid) gives a titer of 1-64 for HCl treated cells; H. mrakii (a predominant diploid) titer of 1-64 for cells treated with ultra violet light; H. schneggii, (an exclusive diploid), the heat treated cells give a titler of 512; H. californica, (a predominant haploid) the U-V treated cells give a titer of 2046; H. conadansis, (a parasitic diploid), both the heat and acid treated cells gave titers of 1024. In all instances, the controls for saline and normal rabbit serum were negative. Some of the treated but unsensitized cells gave good titers with the antibody, thus suggesting that the covering of yeasts are similar to that of some of the Streptococci.

2. Some Carbon Assimilation Studies with Members of the genus Klockers.

Robert Menoser and L. R. Hedrick. Biology Department, Illinois Institute of Technology, Chicago 16, Illinois.

Carbon assimilation studies following the methods suggested by L. Wickerham were performed with several species of the genus Kloeckera. Lodder and Kreger-Van Rij in their new book on yeasts, have made brevis and lindneri synonymous with apiculata. Our studies reveal that brevis certainly should be considered as a separate species, because it can assimilate mannose which K. apiculata cannot, while K. apiculata can assimilate several substances that brevis is unable to, such as, maltose, lactose, galactose, glycerol, melezitose, raffinose, sorbitol, mannetol, inositol, and glucosamine. K.lindneri probably should be a different species as it can assimilate dulcitol and citrate which K. apiculata cannot assimilate.

A key of the <u>Kloeckera</u> yeasts based on their assimilation characters follows:

- 1. (a) dulcitol assimilated 2
 - (b) dulcitol not assimilated 3
- 2. (a) lactose and maltose positive

 i. citrate positive K. lindneri

 ii. citrate negative or weakly positive K. jenseni
 - (b) lactose negative K. corticis

- (a) trohalose asimilated 4 (b) by aclose segetime 5
- μ_{\bullet} (a) maltose, mannose and melezitose assimilated \underline{K}_{\bullet} apiculata
 - (b) melezitose negative, K. magna (c) maltose negative, K. brevis

 - (d) mannosc negative, K africana
- 5. (a) cellobiosc assimilated 6
 - (b) collobiose negative 7
- 6. (a) mannose positive K. javanica
 - (b) mannose negative K. lefari
- 7. (a) mannose and raffinose positive K. antillarum

Yeast work at the Southern Regional Research Laboratory in New Orleans, Louisiana.

Dr. Dorothea J. Tounisson of the Southern Regional Research Laboratory has sent us the following information.

"I have been studying yeasts isolated from rough rice and citrus products. At the present time, I am proparing two manuscripts on the first subject. One will probably be "The Influence of Sealed Bin Storage on the Microbial Populations in Rough Rice" and the other "Yeasts Isolated from Rough Rice Stored in a Scaled Bin."

The letter will include the results of studies on the yeasts done by the methods recommended by Wickerham in the United States Department of Agriculture Technical Bulletin Fumber 1029, 1951. I used all of the carbon compounds he suggested for the assimilation tests, and some others which are constituents of rice. Dr. Mickerham compared the results found with his own data on known strains of yeasts from various sources. He and Mr. Kermit Burton kindly tested some of the assimilation reactions that were doubtful.

Also by these methods, I have re-studied some yeasts isolated from citrus products. I hope to be able to publish one or two papers on this subject. This will include thermal death time data of many of these yeasts in various media. These tests were done by Dr. H. Hall, now of the Northern Regional Research Laboratory, Peoria, Illinois, and myself several years ago."

Studies on Mitrogen metabolism of yeasts.

Dr. P. A. Tetrault has kindly sent the following information concerning activities at the Purdue University, Department of Bacteriology. The up-take and fate of several forms of nitrogen under various conditions is being investigated. The cells are analyzed periodically during growth formetabolic activity, soluble pool nitrogen, lipid-N RNA-N DNA-N, protein N, total lipid and total carboyhdrate. The medium is analyzed for the various nitrogenous substances, pH, turbidity, glucose and phosphate. It is thought that this study will provide basic information for future work by giving a picture of the nutrition and composition of the cells under varying conditions. Methods have been developed for rapid micro and semi-micro total nitrogen and for nitrates in the presence of glucose.

VI. Southern Illinois University.

Gerry Lindegren has sent the following story on activities at this University. It covers the activities of Carl and Gerry Lindegren, and others.

Dr. Maurice Ogur of Brooklyn College, who had formerly spent several short periods of visits to the laboratory, 1950 and in 1951 has joined the staff of the Biological Research Laboratory.

RECENT PUBLICATIONS

- Carl C. Lindegren and Gertrude Lindegren. "Proximity of Genes Controlling the Fermentation of Similar Carbohydrates in Saccharomyces." Nature, Volume 170, page 965, December 6, 1952.
- Carl C. Lindegren and Gertrude Lindegren. "Linkage Relations in Saccharomyces of Genes Controlling the Fermentation of Carboyhdrates and the Synthesis of Vitamins, Amino Acids and Nucleic Acid Components." Indian Phytopathology, Volume IV, Number 1, page 11-20, 1951.

Alvin Sarachek and G. Fred Townsend. "The Disruption of Mitochondria of Saccharomyces by Ultraviolet Irradiation." Science Volume 117, pages 31-33. January 9, 1953.

Carl ^C. Lindegren and Gertrude Lindegren, "Asci in <u>Sac</u>-charomyces with more than four spores". <u>Genetics</u>, Volume 38, pages 73-78, January, 1953.

Norberto J. Pelleroni and Carl C. Lindegren, "A single adaptive enzyme in Saccharomyces elicited by several related substrates". Journal of Bacteriology, Volume 65, pages 122-130, February, 1953.

Carl C. Lindegren. Concepts of Gene-structure and Gene-Action Derived from Tetrad Analysis of Saccharomyces. Experientia Volume 9, pages 75-80, 1953.

VII. University of Pennsylvania, School of Medicine, Department of Dermatology and Syphiology. Dr. Edward D. DeLamater, M.D. Research Professor, has sent the following information:

Dr. E. D. DeLamater, Research Professor of Microbiology, and Research Professor of Dermatology, School of Medicine, has been appointed Director of a new section entitled, section on Cytology and Cytochemistry of Microorganisms, School of Jedicine, University Pennsylvania. He has also just been appointed as a John Simon Guggenheim Fellow for the year 1953-195h, which will permit him to attend the International Congress of Genetics and the International Congress of Microbiology in Bellagio and Rome, Italy, respectively as a guest speaker, and subsequently to spend a period of approximately three months studying in selected European laboratories.

Dr. Mary Elizabeth Hunter of the Section on Cytology and Cytochemistry of Microorganisms, School of Medicine, University of Pennsylvania, will present a paper to the International Congress of Genetics at Bellagio, Rome, on "Meiotic Phenomena in Micrococcus cryophilus".

Mr. Abraham Widra, a student of Dr. DeLamater, is in the process of preparing a paper on "Meiosis in Schizosaccharomyces octosporus".

Mr. Moselio Schaechter, a student of Dr. DeLamater, is currently studying the cytology of Chlamydomonas, and is preparing communications on two aspects of his work which deals with a comparative study of mitosis and chromosome numbers in different species, and the meiotic processes in Chlamydomonas.

Dr, DeLamater is continuing his studies on cytology of meiosis in Saccharomyces cerevisiae, and recently in conjunction with Dr. M. E. Hunter, Dr. V. Bryson, and Dr. W. Szybalski, has demonstrated the influence of terramycin, aureomycin and other antibiotics in producing mitotic arrest. Details of this work will be presented in Bellagio and Rome.

VIII. News from Cecil G. Dunn at Massachusetts Institute of Technology.

The Department of Food Technology, M.I.T. has developed a curriculum in Biochemical Engineering during the last three years. This is the outgrowth of courses in Industrial Microbiology and other subjects given in the Department and at M.I.T. over a long period of years. New courses in Biochemical Engineering are being offered this fall. S.B., S.M., or Ph.D. degrees are now possible in this field.

The Samuel C. Prescott Laboratories was dedicated on June 25. Interested individuals were cordially invited to attend the ceremonies which were held in the afternoon.

Dr. Rugh Burke and associates of the Department of Bacter-iology and Immunology, Harvard Mycology Laboratory, Harvard Medical School, and Boston, have recently isolated an antibiotic in crystalline form from a soil actinomycete which possesses activity towards pathogenic yeasts and other fungi. A report on this research is expected to be published soon.

IV. Current work at the Northern Regional Research Laboratory, Fermentation Division, Culture Collection, Zymology Subsection, News from Dr. Lynferd Vickerham.

Three related genera of ascosporogenous yeasts associated with trees and under investigation. One genus is very weak in its sexual reactions, and some species now placed in Candida belong to it. The second genus is Hansenula which contains heterothallic haploid species. So far as we know, hybridization does not occur between species of Hansenula, but does occur between physiologically different varieties within a species. The third genus of yeasts associated with trees reacily produces hybrids even between its most primitive and most recently evolved species.

Simple procedures that do not require a micromanipulator are used for the isolation of mating types for hybridization and for isolation of haploid hybrids. It is expected that the procedure for the isolation of mating types from diploid yeasts will be presented in a annual meeting of the Mycological Society this fall.

Reprints concerning yeasts received at the N.R.R.L.

They are included because they occurred in Journals which many zymologists may not see regularly.

Stodola, F. H., Odette E. Shotwell, and L. B. Lockwood. 1952. Zymonic acid; a new metabolic product of some yeasts grown in zerated culture. I. Structure studies. J. Am. Chem. Soc., 74: 5415-5418.

Andreason, A. A., and T. J. B. Stier. 1953. Anserobic nutrition of Saccharomyces cerevisiae. I. Ergosterol requirement for growth in a defined medium, J. Cell. and Comp. Physiol., 41: 23-36.

Marquez. 1951. El Aprovechamiento Industrial de los Residuos Agricolas. I. Estudie de la Utilización de los Prehidrolización de Carozos de Maiz (Mazorcas desgranadas) en la Fabricación de Levaduras Alimenticias. Consejo superior de Investigaciónes Cientificas, Cuaderno 1. 50 pages.

Fernandez-Cano, L. H., and J. G. Marquez. 1951. El Aprovechamiento Industrial de les Risiduous Agricolas. 2. Contribucion al Estudio del Metabolismo Carbonado de la Torulopsis utilis Sobre Prehidrolizados de Carozos de siz (Mazorcas desgranadas). Consejo Superior de Investigaciones Cientificas, Cuaderno 2. 45 pages.

Santa Maria, J. 1953. Empleo de los Medios Sinteticos de Wickerham en los Estudios Sobre Nutricion de los Microorganismos. Microbiologia Española, 6: 39-61, with 40 photographs.

Van Uden, N. 1952. Zur Kemtnis von <u>Torulopsis pintolopesii</u> sp. nov. <u>Mit Beobachtungen uber die Parasitare Phase von Acladium castellanii Penoy</u>. Archiv fur Mikrobiologie, <u>17</u>: 199-208.

Zenitani, B. 1952. Yeast occurring in fishery-fermentation products. Part 1. On the generic classification of true yeast in "Shiokara." Science bulletin of the faculty of agriculture, Kyushu University, 12: 21:7-253. Part 2. On the general features of true yeasts in various kinds of "Shiokara." Ibid. 12: 375-381 (1952). Part 3. On the physiological characteristics of true yeasts in "Shiokara." Ibid., 12: 383-390 (1952). English summaries.

Verona, O. 1952. Azione sui lieviti e, in particolare, sui lieviti della formentezione vinario, della vitamine K3eK5. Atti dell'Accademia Italina della Vite e del Viño, 4: 1-15.

Verona, O. 1952. Intorno all'azione esercitata Dagli Insetticidi Sistemici Sulla Microflora del Terreno. L'Agricolturia Italina, 52 (7 n.s.): 61-70.

Tubaki, K. 1952. Studies on the Sporobolobycetaceae in Japan. I. On Tilletiopsis. Nagaoa, issue I, Page 27. II. On Itersonilia, Nagaoa, issue II, Pages 62-66 (1952).

Open House at Lake States Yeast Company.

On May 14, 1953, the Lake States Yeast Company at Rhinelander, Wisconsin, had open house for persons who have been particularly interested in the development of their plant for the conversion of sugars in sulfite waste liquors to cells of Torulopsis utilis to be sold as feed or food yeast. About 150,000 gallons of sulfite liquor per day from the Rhinelander Paper Company flows continuously into a Waldhof propagator. A stream of liquor containing yeast continuously passes from the propagator to the centrifuges, and after washing, goes to the drum driers. The propagator is 26 feet in diameter, contains a central cylinder or draft tube 45 inches in diameter, and a rotating aeration wheel that delivers air just outside the draft tube. The air is very efficiently broken up into tiny bubbles which converts the liquid into a uniform foam. The foam rises outside the draft tube, plunges down through the draft tube, and rises again as it receives fresh air from the aerator wheel. Aqueous ammonia and diammonium phosphate are constantly fed into the Waldhof. The retention time of the sulfite liquor in the propagator is 3.7 hours. The yeast is constantly removed from the Walchef to conical form looker, and thence to separatore. The yeast cream is washed one to three times depending upon the intended use of the yeast. The yeast cream goes to drum dryers and them to flakers. The product has an attractive yellowish color. Five to seven tons are produced daily. The liquor from the first separator contains about 10 percent of calcium lignosulfonates. This liquor is evaporated and the dry powder is sold under the name Toralig. It is used in the

cement industry in oil well drilling procedures and in adhesives. Additional information on this interesting plant may be obtained from a paper in Industrial and Engineering Chemistry, August, 1951.

Officials of the Lake States Yeast Company, the Rhinelander Paper Company, and the Sulphite Pulp Manufacturers' Research League were very gracious hosts.

X. Food Fermentation Investigations Laboratory at North Carolina State College, Raleigh, North Carolina.

The following information has been sent to us by Dr. Paul R. Dawson in behalf of Dr. John Etchells of the Food Fermentation Investigations Laboratory.

Studies on the nature of the yeast flora present on the growing cucumber plant (blossoms, ovaries and fruit) are in their second year. Over 1500 cultures have been isolated and are being identified. The pigmented yeasts were the types most commonly obtained.

The above work has been expanded to cover population changes with respect to species of higher fungi (molds) associated with the cucumber plant throughout the harvest in commercial fields. Over 1,000 cultures from the 1952 season are now being identified in cooperation with mycologists at Harvard University.

An investigation by the Bureau of Agricultural and Industrial Chemistry in cooperation with the Chemistry Department of North Carolina State College has been concerned with the extraction of carotenoid pigments from yeasts. It was observed that the composition of the culture medium greatly influenced the extractability of the pigments from the yeast cells. It is planned to report on this study at the S.A.B. meeting this August in San Francisco, California. Work has now been completed on factors influencing the production of pectin-splitting enzymes from certain yeasts. It was observed that different strains of the same yeast species could vary as much as 50% with respect to enzyme production. This work will be published in the near future.

In the next issue of the botanical journal, Farlowia (Vol. 4, No. 3, 1953) we expect to publish a 40-page article dealing with the morphology of species of yeasts in 12 genera and which should be of considerable interest to the students, teachers, and research workers in the yeast field. The illustrative material dealing with colonial and cellular morphology consists of the following: 12 pages in natural color (15 plates) of yeast colonies and related subjects; 22 pages in black and white (109 plates) mostly of photomicrographs of yeast cells from different cultured media; and 6 pages of text. We expect to have a sufficient number of reprints to meet reasonable demands. The article is entitled "Morphology of Certain Brine Yeasts".

Recent yeast publications:

1. Sugar and acid tolerance of spoilage yeasts from sweet cucumber pickles.

T. A. Bell and J. L. Etchells. Food Technology, 6: 468-472. 1952

- 2. Identification of yeasts from commercial cucumber fermentations in Northern Brining Areas. J. L. Etchells, R. N. Costilow, and T. A. Bell. Farlowia, <u>H</u>: 249-264. 1952.
- 3. Yeasts: Brigands in Brine. J. L. Etchells, T. A. Bell, and I.D. Jones. Research Farming, 10: No. 1, 3-4. 1951.
- 4. Morphology of cortain brine yeasts. J. L. Etchells, T. A. Bell, and I.D. Jones. Farlowia, 4, No. 3, 1953. (In press).
- XI. University of Illinois, Department of Bacteriology, Dr. F. M. Clark has sent the following news.

Marvin P. Steinberg has completed his doctoral research on fet formation by Rhodotorula gracilis. The work was done in the Department of Food Technology under the direction of Professor Z. John Ordal. Mr. Clifford Spatholz plans further work in this field.

In these experiments, an effort was made to study the effect of several variables on the fattening phase as separate from yeast growth. Both fermentor and shake flasks were employed. The fermentor assembly included an automatic pH recorder—controller. The fat content of the yeast was expressed as a ration between yeast fat and non-fat yeast. This fat ratio showed a linear relationship with time. The slope of such a straight line was a measure of the rate of fattening and varied with pH and temperature.

The fat rate varied linearly with pH between pH 3.0 and 8.5 and increased from 2.1 to 3.1 grams fat per 100 grams non-fat dry yeast per hours, respectively. Lowering the temperature of fattening from 28°C. to 22°C. caused a marked decrease in the fat rate, while increasing it to 35°C. decreased the rate only slightly.

Under the experimental conditions used, addition of bios factors to the growth medium was not a prerequisite to subsequent fat formation. This was also true of the cations Ca, Ma, and Mg. Neither sugar concentration up to 16 percent by weight of medium nor hexose used as substrated had a significant effect on fattening.

The maximum fat rate found was greater than given by calculations of data reported by others.

XII. Letter from De Cunha from London. (We sent a note to Dr. De Cunha in Brazil requesting information. Apparently his wife sent it on to him in I ondon and the following letter was forthcoming.)

Today I received a letter from Brazil. The Genetics Congress will be held at the end of August, but I came to Europe two months early to visit the main genetics laboratories in the United Kingdom, France, Switzerland, and Italy. Unfortunately, I cannot send you news about the yeast work. The data I have is in Prazil.

In the last two years, they've been working much more in Drosophila then in yeast problems. They now have three Drosophila articles in print and one in preparation, and only one on yeast and Drosophila. The first mentioned is in collaboration with Dobshansky and will be ready only at about the end of October, since we will be in Europe most of the time. It is about differential attraction with Brazilian yeasts. That is important is that in some places the flies are able to distinguish different strains of the same yeast species.

I have now about 350 strains of yeast to classify. El Tabey will go to Brazil at the end of September and will stay there for fifteen months. Probably very scon we will then have good data on the Brazilian yeasts.

As soon as they have copies of the yeast Drosophila paper, I will send it to you.

ERIEF NOTES

John H. Street, Detroit, Michigan, writes that her only news is the recent paper "A Cytochrome Deficient Mutant of Saccharomyces cerevisiae", which appeared in Experimental and Cellular Research 4, p. 275-305 (1953).

Dr. Milo D. Appleman, Chairman Department of Bacteriology University of Southern California, writes that one of his graduate students has been working on the purification of yeasts from bacteria by means of chemotherapeutic agents. He is working with a rather large group of beer speilage organisms.

Dr. R. C. Hatfield, has distressed us by indicating that he has not received a copy of the Yeast newsletter yet. He was located at the California State Poly Technical College, San Luis Obispo, California, but is now with the Facilities Operation Division, Braddock Heights, Maryland. We hope this copy will reach him.

II. Activities on the Home Ground.

Phaff and Bemain are still working on Pectin breakdown by enzymes obtained from <u>Saccharomyces fragilis</u>. They are gradually pulling the molecules apart, therefor obtaining the mono-dye triand tetra galactronic fractions. Some of this work will be published soon.

Moshe Shrifine completed his master's thesis with Phaff on yeast occurring in the Pine Bark Reetle.

We are also concerned with yeast occurring in green olive brines and on fresh and dried prunes. These projects are presently under way. The only publication has been that by Martin Miller and E.M.Mrak entitled, "Yeasts Associatied with Dried-Fruit Beetles in Figs", Applied Microbiology (7-53)

THE WEXT ISSUE OF YEAST NEWSLETTER

Since I will be away, for the next six or seven months, covering the northern, eastern, and southern fringes of the United States before coming home, Dr. Phaff has agreed to take over editing the letter. Furthermore, when the newsletter was first planned, it was generally agreed that the editorship would alternate. This, however, was interrupted when Dr. Wickerham had his accident. Dr. Phaff, therefore, will take over until Dr. Wickerham is prepared to handle the letter. Should you have any news to include, it would be appreciated if you will send it to Dr. Phaff, so that another issue might come out at the end of the year or the early part of 1954.

S.A.B. Meeting in San Francisco. I neglected to add that many of the yeast fraternity attended the S.A.B.Meeting in San Francisco. Among those that immediately come to mind were Dr. Rudert of the Red Star Yeast Company, Etchells of Raleigh, North Carolina, Howard Douglas from the University of Washington, Dr. Skinner of Washington State College, Leslie Hedrick of the Illinois Institute of Technology, Dr. O.B.Williams of the University of Texas, and others. Most of these men presented papers at the meeting.