

Y E A S T

A News Letter for Persons Interested in Yeast

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	<u>Page</u>
D. Yarrow, Delft, The Netherlands	40
H. J. Phaff, Davis, California	43
S. P. Meyers, Baton Rouge, Louisiana	46
H. Mori, Chiba-ken, Japan	48
Chr. Schönborn, Leipzig, Germany	48
S. Windisch, Berlin, Germany	49
Y. Oshima, Osaka, Japan	51
J. Johnston, Mexico 14, D. F.	52
T. Takahashi, Suita, Japan	52
A. Maxwell, Menlo Park, California	54
H. C. Birnboim, Chalk River, Ontario	55
K. W. van de Poll, Utrecht, The Netherlands	55
J. O. Lampen, New Brunswick, New Jersey	56
H. Suomalainen, Helsinki, Finland	58
H. Pamir, Ankara, Turkey	59
T. M. Enari, Helsinki, Finland	60
Brief News Items	61
International Meetings and Events	65

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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 1971. A contribution of \$1.00 from those who have not contributed for some time would be appreciated to finance future editions of the News Letter. Many thanks to those who have contributed recently. The Editor extends his warmest wishes to the readers of the News Letter for a productive and happy New Year.

H. J. Phaff

I Centraalbureau voor Schimmelcultures (Netherlands), Yeast Division, Delft, Julianalaan 67a. Communicated by D. Yarrow.

Below follows a list of species received by the CBS and which have not been described in the new edition (1970) of The Yeasts (J. Lodder ed.).

Aessosporon salmonicolor van der Walt

J. P. van der Walt, A. v. Leeuwenhoek 36: 49-55 (1970)

Brettanomyces naardenensis Kolfschoten et Yarrow

G. A. Kolfschoten and D. Yarrow, A. v. Leeuwenhoek 36: 458-460 (1970)

Bullera dendrophila van der Walt et Scott

J. P. van der Walt and D. B. Scott, A. v. Leeuwenhoek 36: 383-387 (1970)

Candida australis Goto, Sugiyama et Iizuka

S. Goto et al., Mycologia 61: 748-774 (1969)

C. benhamii Novák et Vitéz

E. K. Novák and I. Vitéz, Zentr. Bakt., Parasit., Infekt., Hyg. 193: 127-133 (1964)

C. bombi Montrocher

R. Montrocher, Rev. Mycol. 32: 69-92 (1967)

C. chilensis Grinbergs et Yarrow

J. Grinbergs and D. Yarrow, A. v. Leeuwenhoek 36: 143-148 (1970)

C. chiropterorum Grose et Marinkelle

E. S. Grose and C. J. Marinkelle, Mycopath. Mycol. Appl. 36: 225-227 (1968)

C. edax van der Walt et Nel

J. P. van der Walt and E. E. Nel, A. v. Leeuwenhoek 34: 106-108 (1968)

C. guilliermondii var. japonica Sugiyama et Goto

J. Sugiyama and S. Goto, J. Fac. Sci. Univ. Tokyo, Sec. 3, 10: 97-116 (1969)

C. incommunis Ohara, Nonomura et Yamazaki

Y. Ohara et al., J. Gen. Appl. Microbiol. 11: 273-275 (1965)

C. ishiwadae Sugiyama et Goto

J. Sugiyama and S. Goto, J. Fac. Sci. Univ. Tokyo, Sec. 3, 10: 97-106 (1969)

C. kosuensis nom. nud.

I. Yokotsuka and S. Goto, J. Agr. Chem. Soc. Japan 29: 132-135 (1955)  
= C. boidinii, J. Grinbergs and D. Yarrow, A. v. Leeuwenhoek 36: 143-148 (1970)

C. oleophila Montrocher

R. Montrocher, Rev. Mycol. 32: 69-92 (1967). The strain, CBS 2219, indicated as the type was identified as C. sake by N. van Uden and H. Buckley, The Yeasts, 1970, p. 1038.

Candida parapsilosis v. hokkai nom. nud.

S. Goto and I. Yokotsuka, Bull. R. Inst. Ferm. Yamanashi Univ. 9: 79-87 (1962)

C. punicea Komagata et Nakase

K. Komagata and T. Nakase, J. Gen. Appl. Microbiol. 11: 255-267 (1965)  
= Sporobolomyces sp., D. Yarrow, A. v. Leeuwenhoek 35: 24-28 (1969)

C. requinyii Szép et Novák

E. Szép and E. K. Novák, Acta Bot. Acad. Sci. Hung. 9: 447-453 (1963)

C. steatolytica Yarrow

D. Yarrow, A. v. Leeuwenhoek 35: 24-28 (1969)

C. suecica Miranda et Norkrans

L. R. de Miranda and B. Norkrans, A. v. Leeuwenhoek 34: 115-118 (1968)

C. tepae Grinbergs

J. Grinbergs, Arch. Mikrobiol. 56: 202-204 (1967)

C. terebra Sugiyama et Goto

J. Sugiyama et S. Goto, J. Fac. Sci. Univ. Tokyo, Sec. 3, 10: 97-116 (1969)

C. valdiviana Grinbergs et Yarrow

J. Grinbergs and D. Yarrow, A. v. Leeuwenhoek 36: 143-148 (1970)

Debaryomyces halotolerans nom. nud.

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

D. nepalensis Sugiyama et Goto

S. Goto and J. Sugiyama, J. Jap. Bot. 43: 102-108 (1968)

Endomycopsis lipolytica Wickerham, Kurtzman et Herman

L. J. Wickerham et al., Science 167: 1141 (1970)

E. muscicola Nakase et Komagata

T. Nakase and K. Komagata, J. Gen. Appl. Microbiol. 12: 347-352 (1966)

Hansenula philodendra Scott et van der Walt

D. B. Scott and J. P. van der Walt, A. v. Leeuwenhoek 36: 389-396 (1970)

H. sydowiorum Scott et van der Walt

loc. cit.

Pichia castillae Santa Maria et Aser

J. Santa Maria y C. G. Aser, Bol. Inst. Nac. Invest. Agronom. 62: 51-55 (1970)

P. krusei nom. nud.

T. Tsuchiya, Y. Fukazawa, T. Shinoda and M. Imai, Japan. J. Exp. Med. 37: 285-290 (1967)

Rhodotorula araucariae Grinbergs et Yarrow

J. Grinbergs and D. Yarrow, A. v. Leeuwenhoek 36: 455-457 (1970)

- Rhodotorula glutinis var. rufusa Iizuka et Goto  
H. Iizuka and S. Goto, J. Gen. Appl. Microbiol. 11: 331-337 (1965)
- Saccharomyces beticus Marcilla, Alas et Feduchy ex Santa Maria  
J. Santa Maria, Inst. Nac. Invest. Agronom. 62: 57-66 (1970)
- S. cordubensis Santa Maria  
loc. cit.
- S. gaditensis Santa Maria  
loc. cit.
- Selenotila intestinalis Krassilnikov  
N. A. Krassilnikov, Microbiological Journal (U.S.S.R.) 4: 134-137  
(1927), D. Yarrow, A. v. Leeuwenhoek 35: 418-420 (1969)
- Sel. peltata Yarrow  
D. Yarrow, A. v. Leeuwenhoek 35: 418-420 (1969)
- Sporobolomyces antarcticus Goto, Sugiyama et Iizuka  
S. Goto et al., Mycologia 61: 748-774 (1969)
- Sterigmatomyces elviae Sonck et Yarrow  
C. E. Sonck and D. Yarrow, A. v. Leeuwenhoek 35: 172-177 (1969)
- St. polyborus Scott et van der Walt  
D. B. Scott and J. P. van der Walt, A. v. Leeuwenhoek 36: 389-396  
(1970)
- Syringospora albicans (Robin) Dodge  
J. P. van der Walt, Mycopath. Mycol. Appl. 40: 231-243 (1970)
- Syr. claussenii van der Walt  
loc. cit.
- Syr. stellatoidea van der Walt  
loc. cit.
- Torulopsis bombicola Spencer, Gorin et Tulloch  
J. F. T. Spencer et al., A. v. Leeuwenhoek 36: 129-133 (1970)
- T. humilis Nel et van der Walt  
E. E. Nel and J. P. van der Walt, Mycopath. Mycol. Appl. 36: 94-96  
(1968)
- T. karawaiewi nom. nud.  
G. Jurzitza, Arch. Mikrobiol. 72: 203-222 (1970)
- T. kestonii Scarr et Rose  
M. P. Scarr and D. Rose, J. gen. Microbiol. 45: 9-16 (1966)
- T. mannitofaciens Onishi et Suzuki  
H. Onishi and T. Suzuki, A. v. Leeuwenhoek 35: 258-260 (1969)
- T. psychrophila Goto, Sugiyama et Iizuka  
S. Goto et al., Mycologia 61: 748-774 (1969)

Torulopsis xestobii nom. nud.

G. Jurzitza, Arch. Mikrobiol. 72: 203-222 (1970)

Trichosporon aquatile Hedrick et Dupont

L. R. Hedrick and P. D. Dupont, A. v. Leeuwenhoek 34: 474-482 (1968)

Tr. cutaneum var. antarcticum Goto, Sugiyama et Iizuka

S. Goto et al., Mycologia 61: 748-774 (1969)

Tr. eriense Hedrick et Dupont

L. R. Hedrick and P. D. Dupont, A. v. Leeuwenhoek 34: 474-482 (1968)

Tr. fennicum Sonck et Yarrow

C. E. Sonck and D. Yarrow, A. v. Leeuwenhoek 35: 172-177 (1969)

Tr. melibiosaceum Scott et van der Walt

D. B. Scott and J. P. van der Walt, A. v. Leeuwenhoek 36: 389-396 (1970)

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

1. Below follow two abstracts of articles by Professor M. W. Miller and coworkers.

ENDOMYCES TETRASPERMA, A NEW SPECIES

J. M. MACY AND M. W. MILLER

Department of Food Science and Technology  
University of California, Davis, Calif., U.S.A. 95616

MACY, J. M. AND M. W. MILLER. 197-. Endomyces tetrasperma, A New Species. Journal of Bacteriology (in press).

A new fungal species has been described and placed in the genus Endomyces. Endomyces tetrasperma forms a true septate, multinucleate mycelium which breaks up into arthrospores. Ascus formation occurs after isogamous copulation between sexual protuberances which develop at the ends of arthrospores or between two cells, adjacent mycelial cells or arthrospores. The asci which dehisce at maturity release 2-4 smooth, ovoid, thick walled spores, each containing two oil droplets. The proposed life cycle is based on morphological and cytological observations.

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Synonymy of Metschnikowia pulcherrima and  
Torulopsis burgeffiana

M. W. Miller, Ellen R. Johnson and J. I. Pitt\*

Department of Food Science and Technology  
University of California, Davis, Calif., U.S.A. 95616

Journal of Bacteriology (in press)

Abstract

Torulopsis burgeffiana is placed in the genus Metschnikowia because it has been shown to have a perfect stage corresponding with this genus. Due to the characteristic shape of the ascus and ascospores, T. burgeffiana is to be considered a synonym of M. pulcherrima. The physiological differences which exist between strains are not sufficiently important to maintain a separate species.

Benda (1962) described Torulopsis burgeffiana when she compared her isolates with Candida pulcherrima and concluded that the differences were such that her isolates belonged to a new species.

In the course of our studies on the genus Metschnikowia, C. pulcherrima was induced to form ascospores and subsequently transferred to the genus Metschnikowia (Pitt and Miller, 1968).

As the T. burgeffiana isolates had many characteristics similar to M. pulcherrima, including the formation of pulcherrima cells (Fig. 1), subcultures of three of Benda's isolates were obtained from van Uden (Gulbenkian Institute, Lisbon, Portugal) and the ability to form ascospores was re-examined. Sphaeropedunculate asci containing 2 acicular ascospores per ascus (Fig. 2) were formed after two to three weeks at 12 C. on a dilute (1:99) filtered V-8 juice agar medium described by Pitt and Miller (1968). The number and shape of the ascospores was determined by micromanipulation.

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Australia.

2. The following is an abstract of the Ph.D. dissertation by Dr. Sally A. Meyer. The work was done under the guidance of Prof. H. J. Phaff.

DNA Base Composition and Homology in Candida Species  
and Related Yeasts

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Department of Food Science and Technology  
University of California, Davis, Calif., U.S.A. 95616

Abstract

The DNA base composition was determined for 55 strains of Candida representing 30 species and four varieties. The range of GC values was 33 to 57.3 moles %. Twelve ascosporeogenous yeasts belonging to the genera Lodderomyces and Metschnikowia were also studied and these showed a range of 39.5 to 49.0 moles % GC. The GC contents for DNA of members of the Candida parapsilosis group sensu Phaff et do Carmo-Sousa ranged from 40.5 to 48.1 % GC, while the range for the expanded C. parapsilosis group sensu Fell et Meyer was from 35.9 to 57.3 % GC.

DNA-DNA hybridization experiments indicated no significant degree of homology between C. parapsilosis and any other species tested in this study. These included strains which have been considered varieties (C. parapsilosis var. querci and C. parapsilosis var. intermedia), and L. elongisporus, the proposed perfect stage of C. parapsilosis. Both varieties appear distinct from C. parapsilosis and should be considered as separate species. The proposal that L. elongisporus is the perfect stage of C. parapsilosis is rejected.

High degrees of DNA homology were shown between C. salmonicola and C. sake as well as between C. lusitaniae and C. obtusa. It is proposed that these two pairs of species be reduced to two species, viz. C. sake and C. lusitaniae, respectively.

Five strains of C. guilliermondii var. guilliermondii had GC values of 45.1 to 46.3 %. High degrees of DNA homology between the type strain and the four other strains were demonstrated at 25C below the  $T_m$ , but at 14C below the  $T_m$  three of the strains revealed some reduction in % homology. Likewise C. guilliermondii var. carphophila showed a high degree of DNA homology with the variety guilliermondii.

C. pulcherrima and M. pulcherrima (which are represented by different type strains) demonstrated a high degree of DNA homology with each other. No other Metschnikowia species showed a significant degree of DNA homology with M. pulcherrima.

Based on GC % and overall physiological and morphological properties, the following pairs of species appear to be closely related: C. brumptii and C. ravautii, C. aaseri and C. tenuis, and C. tropicalis and C. arborea.

Eight strains of C. diddensii (including the type strain of C. atmosphaerica) and C. polymorpha (type strain) showed a range of GC contents from 36.6 to 39.3 %. These nine strains may represent either a single heterogeneous species or two separate species which can not be differentiated by currently used physiological criteria.

The GC content of C. viswanathii is ca. 10 % higher than that of C. albicans or C. tropicalis. A close relationship to either of these last two species is not supported. C. bogoriensis has 57.3 % GC, the highest of any strain included in the entire study of 67 yeasts. This organism is distinctly different from any of the other members of the C. parapsilosis group. Its high GC content and other properties suggest a relationship with the heterobasidiomycetous yeasts.

The so-called C. parapsilosis groups sensu Phaff et do Carmo-Sousa and sensu Fell et Meyer are heterogeneous groups of what appear to be mostly unrelated organisms. In general, the physiological similarities of the organisms in the group are not indicative of a high degree of genetic relatedness. It appears that in many instances physiological properties used in current systematics are not adequate for gathering species into groups, or, possibly even strains into species. Separation of species on the basis of a single assimilative or fermentative property is unsound. It is suggested that GC values, and preferably DNA homology, should be included in future taxonomic studies before new organisms are described and named or new relationships are proposed.

III Louisiana State University, Baton Rouge, Louisiana 70803. Department of Food Science. Communicated by S. P. Meyers.

Recent publication: Mycological studies of Lake Champlain. Mycologia 62: 504-515. 1970 (with D. G. Ahearn and W. L. Cook).

Dr. Meyers and D. G. Ahearn (Georgia State University, Atlanta, Ga.) have established a cooperative research program on hydrocarbonoclastic yeasts with emphasis on those from aquatic localities. The following manuscripts have been prepared in this subject area:



The role of yeasts in the decomposition of oils in marine environments. (D. G. Ahearn, S. P. Meyers and P. G. Standard) Dev. in Industrial Microbiology 12: 000-000. 1971. (Presented at the annual meeting of the Soc. for Indust. Microbiology, Kingston, Rhode Island, August, 1970)

ABSTRACT

Yeasts from various marine, fresh water and terrestrial environments were examined for their capacity to assimilate hydrocarbons. Hydrocarbonoclastic yeasts are widespread, however, strains able to assimilate hydrocarbons at levels > 2% (v/v) generally are concentrated in oil-polluted habitats. Louisiana crude oil and refinery and laboratory fractions, and vapors of a number of aromatic hydrocarbons, served as effective carbon sources for growth of selected isolates. Most organisms showed yeast-like growth on the surface of the hydrocarbon globules, but certain strains, mainly marine isolates of Trichosporon cutaneum, penetrated and developed within the globule. Field and laboratory data suggest that yeasts play a role in microbial decomposition of surface oil depositions in the marine environment.

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Mycological degradation of petroleum products in marine environments. (S. P. Meyers and D. G. Ahearn). FAO Technical Conference on Marine Pollution and its Effects on Living Resources and Fishing. Rome, Italy, 1970

ABSTRACT

Yeasts and yeast-like fungi are widespread in estuarine and marine waters, but rarely exceed 500 viable units/100 ml. Significantly increased densities are observed in organically-enriched habitats and those subject to pollution by petroleum and petroleum products. In coastal marsh sediments, yeast concentrations as great as  $9 \times 10^4$  viable units/cm<sup>3</sup> of sediment are encountered. Pollution of waters or beaches with petroleum may result in significant alteration of the composition of the normal yeast biota, with concurrent increase in numbers of hydrocarbonoclastic species. These latter utilize a range of hydrocarbons including alkanes from C<sub>10</sub>-C<sub>18</sub>, kerosene, diesel fuel, and tractor fuel. Respiration of mixed populations of selected yeasts grown on crude oil fractions is greater than that obtained with individual organisms alone. The use of a selected combination of yeasts in biodegradation of oil spills is postulated, employing carefully designated delivery systems using micro-encapsulation principles.

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Dr. Meyers has recently received support from the Federal Water Quality Control Administration to examine aspects of microbial mediation of oil in the aquatic environment, with particular attention given to the productive region of the Louisiana marshland. Tests are proposed to evaluate hydrocarbon utilization by marine-occurring yeasts and yeast-like fungi in this area and to ascertain the response of these organisms to oil intrusion into the marshland rhizosphere. Attention will be given to ultimate development of techniques for accelerated biodegradation of oil pollutants in such aquatic habitats.

Stipends are now available for graduate study in the Departments of Food Science and Marine Science at Louisiana State University (M.S. and Ph.D. degrees). Major emphasis is on mycology and development of food resources in addition to a range of related marine mycological problems. For information contact Dr. Meyers.

IV Noda Institute for Scientific Research, Noda-shi, Chiba-ken, Japan.  
Communicated by Haruhiko Mori.

The following paper was recently submitted for publication to J. Ferment. Technol.

A YEAST ISOLATED FROM TOMATO CATCHUP

H. Mori, S. Nasuno and N. Iguchi

ABSTRACT

A yeast was isolated from tomato catchup spoiled during preservation tests of the products and identified as Saccharomyces acidifaciens (Nickerson). This yeast was vigorously gas-forming and produced large amounts of acetic acid and ethyl alcohol in catchup. The vegetative cells and ascospores were tolerant to wet heating for 5 minutes at 62C and 67C, respectively. It showed strong ability for sporulation, especially on V-8 agar or tomato juice agar, and also exhibited characteristics of haploid yeasts, which, on various sporulation media, form only zygotic asci. Furthermore, this yeast was bisexual and homothallic. However, the large cells, which formed only azygotic asci and which seemed to be diploid, were obtained by trials for breeding of diploid cells, that is, by transfer of a single zygote into a droplet of yeast maintenance medium. In the asci, several azygotic 5 to 10 spored-asci and zygotic 5 to 8 spored-asci were occasionally observed. Manufacturers of tomato catchup need to pay special attention to contamination by S. acidifaciens, which possesses active fermentability and ability for sporulation and its growth may be promoted by tomato juice.

Now, we are studying on genetic improvement of S. rouxii, a commercially important yeast for shoyu and miso fermentation.

V Karl-Marx-Universität, Klinik für Hautkrankheiten, Mykologische Abteilung, Liebigstr. 21, Leipzig, D.D.R. 701. Communicated by Dr. Chr. Schönborn.

To demonstrate Cryptococcus neoformans 357 samples of bird excreta were examined for yeasts. There were 117 samples from wild birds, 147 from pigeons gone wild and 103 from birds of the zoo. The culturing-medium consisted of Guizotia abyssinica-creatine-antibiotic-diphenyl agar (STAIB and SEELIGER).

Yeast fungi were found in 94.9% of all samples examined, but Cryptococcus neoformans was not demonstrated. A total of 762 microorganisms were isolated by culturing, these mainly belonged to the genera Rhodotorula (19.7%), Torulopsis (18.4%), Candida (15.6%), Geotrichum (11.5%). The most frequent fungi were Rhodotorula minuta, Geotrichum candidum, Aureobasidium

spec., Torulopsis famata, Trichosporon pullulans, Candida albicans, Torulopsis glabrata, Kloeckera apiculata and Candida krusei. The yeast flora of wild birds showed the greatest variety of species. Candida albicans was cultured once from pigeon excreta, 7 samples from zoo birds and 14 from wild indigenous birds.

A comparison of the intestinal yeast flora of birds, mammals and man showed that birds do not have a specific faecal flora, but that apparently conditions for species of the genera Debaryomyces, Rhodotorula, Kloeckera and Cryptococcus are more favourable.

For nearly all microorganisms isolated, a habitat outside the homoiothermic organism, in the direct environment of the birds, has been found.

In addition to Candida albicans the following yeasts facultatively pathogenic to man were found: Candida pseudotropicalis (Sacch. fragilis), Candida parapsilosis, Candida krusei (Pichia fermentans), Candida guilliermondii and Torulopsis glabrata.

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

Since the last contribution the following publications have appeared:

U. Steckowski: Regulationsmechanismen der  $\alpha$ -Glucosidase-Synthese bei Saccharomyces-Hefen  
Thesis, Technical University Berlin 1969

S. Windisch u. U. Steckowski:

Versuche mit osmotoleranten und Backhefen zum Ersatz chemischer Teiglockerungsmittel.

(Experiments with osmotolerant and baking yeasts as a substitute for chemical leavening agents)

Internatl. Zeitschrift f. Lebensmittel und Lebensmittel-technologie GORDIAN 1970, 336-338, 381 + 382, 434 + 435.

SUMMARY

The usual baking yeasts are not able to take the place of chemical leavening agents in biscuit doughs rich in fat and sugar and poor in water. Therefore one is dependent on the use of chemical leavening agents for biscuit production.

Experiments to breed new types of yeast which are able to act as leavening agents in doughs with low amounts of water and which can replace chemicals have been made by crossing baking yeasts with osmotolerant wild types. They have shown that breeding is possible but rather difficult. By screening yeasts for types fit for crossing experiments strains have been found which show in the fermentometer of Burrows and Harrison sufficient fermentative activity. These strains could be propagated in batch culture. The loss of fermentative activity during growth under strong aeration is compensated by minor changes of physical conditions. Large scale baking experiments resulted in

products which suggest that special osmotolerant yeast strains are possibly a means to compensate for chemical leavening agents. Such yeasts only ferment doughs rich in fat and sugar and poor in water, but not normal wheat flour doughs.

S. Windisch u. I. Neumann:

Hefen in Mandelmassen und Süßwaren.

(Yeasts in ground almond and sweets)

Süßwaren 13, 1406-1410, 1969

SUMMARY

Ground almonds, marzipan and other sweet articles have all a more or less small content of water and a rich one of sugar. However, 193 strains of fermentative yeasts have been isolated from them and determined. They were tested for their ability to ferment solutions with 45, 60, 75 resp. 85% fructose (w/v). Moreover 31 strains of nonfermentative yeasts have been found. The fermenting group contained 27 strains, which could still ferment in solutions with 75% fructose. The most frequent species of the 33 fermentative ones were Saccharomyces rouxii, S. carlsbergensis and S. cerevisiae. S. rouxii is well known as a cause of spoiling marzipan. The properties of the isolated yeasts are discussed, especially the significance of osmotolerance and slime formation for ecology and spoilage.

S. Windisch u. I. Neumann:

Über Resistenz, Bedeutung und Bekämpfung von Saccharomyces carlsbergensis und Saccharomyces rouxii in Marzipan.

(On resistance, importance and control of Saccharomyces carlsbergensis and Saccharomyces rouxii in marzipan)

Proc. 2nd Symposium Technical Microbiology, Berlin 1970, 415-422

SUMMARY

The most frequent yeasts in masses of ground almonds and in marzipan are Saccharomyces rouxii and S. carlsbergensis, the latter well known as the bottom fermenting beer yeast. S.r. is a spoilage organism, which causes bursting of marzipan by CO<sub>2</sub> formation during storage. These yeasts are experimentally compared, because they occur regularly in almond masses. Both yeasts are highly resistant against increased temperature and stand 85°C for more than 40 minutes. No other species found in marzipan is able to withstand such conditions. Equally those two yeasts are the most resistant ones against benzaldehyde. We made experiments with and without benzaldehyde and used different temperatures during times from 1 to 40 minutes. We found that 0.02% benzaldehyde, which does occur in marzipan, affects both yeasts equally and independently from temperature. The only striking difference between the two yeasts seems to be the marked osmotolerance of S.r., though this property does not qualify a species. Obviously S.r. and S.ca. are closely related and neither of them die off in marzipan. However, S.ca. is not able to ferment in marzipan and therefore is no cause of spoilage. Using the results obtained the authors consider the necessity to develop a better technique in the production of rough almond masses.

G. Koppensteiner: Ursachen und Auswirkungen der Osmotoleranz bei Hefen.  
(Causes and effects of osmotolerance with yeasts)  
Thesis, Technical University Berlin 1969

G. Koppensteiner: Der osmotische Wert als Parameter für Wachstum und  
Alkoholbildung bei Hefen.  
(The Osmotic Value as a Parameter of Growth and of Ethanol  
Formation in Yeast)

Z. Naturforsch. 25 b, 606-610, 1970

SUMMARY

The change of the osmotic value of different substances was observed during yeast growth by determination of the lowering of freezing-points. In shaken cultures with fermentable sugars the osmotic value at first increased and then decreased (effect of diauxy). Different attenuation degrees of raffinose (1/3, 3/3) as such reflected the osmotic value of the fermented medium. The diminution of the osmotic value in liquid auxanograms showed distinctly whether the yeast had grown by utilizing the sugar or by using the carbon of amino acids or of reserve substances in the cell itself. By the aid of a calibration curve the ethanol content of a fermented medium may be determined exactly.

VII Osaka University, Higashinoda, Miyakojima, Osaka, Japan. Communicated by Yasuji Oshima.

I have recently moved from the Central Research Institute, Suntory Ltd., to the Department of Fermentation Technology, Faculty of Engineering, Osaka University, Yamada-kami, Suita-shi, Osaka 565, Japan.

One of our main projects, genetic studies on homothallism genes in Saccharomyces, has fairly well progressed. Our present conclusions are as follows:

The a and α mating-type alleles in Saccharomyces are interchangeable with each other by mutagenic action of two unlinked homothallism genes, HO<sub>α</sub> and HM. Conversion from an a to an α requires both HO<sub>α</sub> and HM, while only HO<sub>α</sub> is required for conversion from α to a. Those conversions are performed with extraordinarily high frequency, probably 100%, within a few generations subsequent to spore germination. The mutagenic effects of the homothallism genes for the mating-type locus are strictly locus specific.

Details of these studies have appeared or will be published shortly in the following publications:

I. Takano & Y. Oshima, 1970. Allelism tests among various homothallism controlling genes and gene systems in Saccharomyces. Genetics 64: 229-238.

I. Takano & Y. Oshima, 1970. Mutational nature of an allele specific conversion of the mating type by the homothallic gene HO<sub>α</sub> in Saccharomyces.

Genetics 65: in press.

Y. Oshima & I. Takano. Mating types in *Saccharomyces* yeasts; Their convertibility and homothallism. in preparation.

VIII Centro de Investigacion y de Estudios Avanzados del Instituto Politecnico Nacional, Departamento Genética y Biología Celular, Apartado Postal 14-740, Mexico 14, D. F. Communicated by John Johnston.

I am spending a nine-months sabbatical leave from the University of Strathclyde in the Department of Genetics and Cell Biology of this Institute. I am working on more extensive genetic analysis of polyene antibiotic resistance in yeast. In traveling here it was a great pleasure to visit the Laboratories of Professor J. J. Miller in Hamilton, Ontario and Dr. F. Sherman in Rochester, New York. I am hoping to have an opportunity to visit several Northamerican yeast laboratories during my return journey in June and July. I would be pleased to be contacted by other persons interested in yeast who are either working in or visiting Mexico.

Mr. John Coulson has recently completed his research for his Ph.D. thesis. Resistant mutants isolated from a) amphotericin B, and b) pimarinic, and nystatin-resistant mutants isolated previously have been classified into several groups according to their cross-resistance patterns to five polyene antibiotics (amphotericin B, candicidin, lagosin, nystatin and pimarinic). There is an apparent relationship between cross-resistance pattern and allelism between different mutants. All these mutants were recessive and allelism tests suggest the existence of six or more loci. Tetrad analysis of many inter-mutant, sensitive diploids gives a most interesting result, a 2:2 segregation for resistance v. sensitivity. (Similar results were obtained earlier by Dr. P. V. Patel with dominant mutants.) A model proposing resistance-suppressor genes, acting through a "cytoplasmic factor" which would most likely be the cell membrane, has been examined. An alternative idea, favored by us, is that all resistance mutants are cell-membrane mutants, but some are expressed only in specific residual membrane genotypes. As yet, our crosses have not been between isogenic strains and segregants are therefore likely to have recombinant membrane-structure genotypes. We propose that some resistance mutants are non-specific, being expressed in membrane recombinants while others are specific, recombinant membranes showing polyene sensitivity. This model would also explain many cases of low spore-viability in crosses and a few cases of "conditional lethality", when crosses show 2:2 segregation for lethality. It is possible that the proposed non-specific mutants are ergosterol-deficient whereas "specific mutants" result in more subtle changes of sterol: phospholipid ratio.

IX Suita Laboratory, Brewing Science Research Institute, Asahi Breweries Ltd., 5-3 Deguchi-cho, Suita, Japan. Communicated by T. Takahashi.

The second meeting of Yeast Genetics Conference-Japan was held on August 26 and 27, 1970.

An outline of the meeting follows below.

The second meeting of the Yeast Genetics Conference-Japan was held on August 26 and 27, 1970, at the

Seminar Room, Department of Fermentation Technology, Osaka University. Forty yeast investigators met, and six general areas were discussed: sexuality, mutation and radiation effects, heritable characters, gene action and regulation, cytoplasmic inheritance, and cytology.

Takano and Oshima (Osaka) presented their works on homothallism and heterothallism summarily. The details will be published in the near future. Gunge (Yokohama) found an unusual strain which was characterized by aa and sporulation ability. The strain was isolated as a segregant of triploid strain. Such strain was also isolated by Takano and Oshima. Takahashi (Suita) reported the sporulation negative hybrids between haploid segregants obtained by gene conversion of gene D. Mori (Noda) demonstrated the life cycle of S. rouxii, which has been used for soya sauce production. Unfortunately, genetic analysis of the yeast has not advanced, because mutant induction was very difficult and the crossing between this yeast and S. cerevisiae was impossible.

Nakai (Chiba) was studying the repair mechanism and the difference between mutation, gene conversion and recombination by the radiological studies with use of uvs, xrs and their double mutants. Ito (Tokyo) demonstrated the genetic effects of photodynamic action and radiations.

Takahashi isolated gene controlled congo-red resistant strain by UV irradiation. He also found a phenethyl alcohol resistant strain among his stock cultures, and the presence of at least two PEA genes were supposed.

Takano proposed a new pathway of methionine and threonine biosynthesis, based on the genetic analyses of the hybrids of various met and hom mutants. Tamaki (Kyoto) presented his data on amylase activities of the segregants of the hybrids originated from S. diastaticus and S. cerevisiae. The segregation was not 2 : 2, and a complex regulation mechanism on amylase synthesis was proposed.

Wakabayashi (Tokyo) and Gunge expressed the cytoplasmic inheritance of oligomycin resistance. This character was not linked to rho. Nagai (Nara) reported the high frequency production of RD mutant using wild yeasts in a nutrient deficient medium. Morita (Shizuoka) demonstrated the relation of RD mutation and cell cycles, especially reproduction of mitochondria. Yamamoto (Osaka) showed the difference in r-RNA between strontium-resistant and sensitive strains.

Yuasa (Tokyo) demonstrated pictures on meiosis of various yeasts. Hirano (Tokyo) observed the fine structure of protoplasmic membrane under the electron microscope using freeze-etching method. Osumi (Tokyo) and Sando (Tsuruoka) reported abnormal mitochondria formation by the effect of chloramphenicol and cycloheximide. Sando and Hayashibe (Tsuruoka) analyzed the chemical components of bud scar. The main component was chitin. Iguti (Mito) reported yeast growth in the presence of snail digestive juice.

Finally, Ito introduced the topics on radiation biology during his travelling in USA and Germany.

X Stanford Research Institute, Menlo Park, California 94025. Communicated by A. Maxwell.

Below follows an abstract for a paper which A. Maxwell presented at the Cell Biology meetings in San Diego, November 20, 1970.

Ultrastructural Changes in Saccharomyces cerevisiae  
Treated with Iodoacetic Acid

W. A. Maxwell and Edward Spoerl  
Stanford Research Institute, Menlo Park, Ca. 94025  
and

U. S. Army Medical Research Laboratory, Fort Knox, Kentucky 40121

Iodoacetic acid (IAA) is known to have marked effects on the physiology and biochemistry of living cells and has been used extensively as an inhibitor of energy production. Cytological changes in yeast incubated with IAA have been examined in an attempt to correlate ultrastructural changes with physiological data which indicate that cell membranes are affected by IAA. Cells of Saccharomyces cerevisiae were incubated for 30 min in buffer solutions of IAA ranging from 1 to 10 mM in concentration. Ultrastructural changes are obvious in electron micrographs of thin sections of chemically fixed cells and of samples prepared by freeze etching. At low concentrations (3 mM) vacuoles are most obviously affected as shown by distorted and broken vacuolar membranes. At higher concentrations the vacuoles become partly or wholly filled with cytoplasmic material. As IAA concentrations are increased, changes occur in mitochondria, nuclei, and general cytoplasmic organization. Most of these effects appear first as distortions or alterations in the membranes affected organelles. Alterations of the surface of vacuolar membranes are also shown in freeze etch preparations and are consistent with thin section results. Selective disruption of vacuolar membranes, compared with external membranes, is consistent with the efflux of sugars from yeast treated with IAA. Efflux of sorbose from treated cells occurs at a single uniform rate, in contrast to efflux from normal cells which includes a second slower rate, apparently due to compartmentation of sorbose in the cell vacuole (J. Membrane Biol. 1: 468, 169).



XI Biology and Health Physics Division, Atomic Energy of Canada Limited, Chalk River, Ontario. Communicated by H. C. Birnboim.

NUTRITIONAL REQUIREMENT OF "WILD TYPE"  
SCHIZOSACCHAROMYCES POMBE FOR CYSTEINE AT  
LOW CELL CONCENTRATIONS

In the course of experiments requiring log phase Sch. pombe cells in minimal medium, we found that a wild type strain, 972h<sup>-</sup>, exhibited an unexpectedly long lag in their growth curve if the initial cell concentration was less than 5 - 10 x 10<sup>4</sup> cells/ml. The lag ranged from one day to as long as seven days if the initial cell number was as low as 10<sup>2</sup> - 10<sup>3</sup> cells/ml. Such a pronounced lag did not occur if cells were grown in complete medium, and was less if the minimal medium was supplemented with casamino acids. Individual amino acids were then tested. A slight stimulation was obtained with either arginine, glycine, histidine, or methionine added individually. A marked stimulation, however, was noted when L-CYSTEINE (filter sterilized) was added to a final concentration of 0.02 mg/ml. In this case, the lag phase was significantly reduced and cultures now started at 10<sup>2</sup> cells/ml, routinely grew to 1 - 5 x 10<sup>6</sup> cells/ml over a weekend. In addition to cysteine, the medium used contained the following organic components: asparagine, calcium pantothenate, nicotinic acid, myo-inositol, biotin, and 0.5% fructose as a carbon source. The pH was buffered at 5.5 with sodium acetate.

XII Van't Hoff Laboratory, State University of Utrecht, Sterrenbos 19, Utrecht, The Netherlands. Communicated by K. W. van de Poll.

Recently the following thesis was completed:

Regulation of allantoinase synthesis in  
Saccharomyces carlsbergensis

K. W. van de Poll

Allantoinase synthesis, like the synthesis of other enzymes in yeast is repressable by a nitrogen source, most actively by ammonium ions, asparagine and glutamine; the latter two probably produce ammonium ions. On the other hand, glucose exerts a specific stimulatory effect on derepressed allantoinase synthesis. Growth experiments indicated that the nature of the carbon source primarily determines the degree of derepression.

$\alpha$ -Glucosidase synthesis, like the synthesis of many other enzymes in yeast, is subjected to catabolite repression. Moreover, ammonium ions, asparagine and glutamine exert a specific stimulatory effect on the derepressed synthesis of this enzyme. It is generally assumed that the effector of catabolite repression is an intermediate of carbon metabolism. The same assumption was made now for allantoinase synthesis. Thus, ammonium ions might affect the synthesis of both enzymes via carbon metabolism. Indeed, it appeared that ammonium ions profoundly influence the activity of the HMP shunt and the reserve carbohydrate synthesis under conditions optimal for  $\alpha$ -glucosidase, and both these pathways and the

glycolysis under conditions optimal for allantoinase synthesis. Although no definite conclusions can be made about the nature of the effector(s) of  $\alpha$ -glucosidase and of allantoinase synthesis, many intermediates of the carbon metabolism could be excluded.

These results were published in the following papers:

- K. W. van de Poll, A. A. G. Verwey and V. V. Koningsberger, Proc. Kon. Ned. Akad. Wet. B71 (1968) 344
- K. W. van de Poll, Proc. Kon. Ned. Akad. Wet. B73 (1970) 10
- K. W. van de Poll, Proc. Kon. Ned. Akad. Wet. B73 (1970) 342
- R. van Wijk, K. W. van de Poll and G. A. G. Speziali, Proc. Kon. Ned. Akad. Wet. B73 (1970) 357
- K. W. van de Poll and R. van Wijk, Proc. Kon. Ned. Akad. Wet. B73 (1970) 372.

XIII Rutgers University, The State University of New Jersey, Institute of Microbiology, New Brunswick, New Jersey 08903. Communicated by J. O. Lampen.

Five papers related to yeast have appeared or are in press from this laboratory during the past year.

- (1) Sentandreu, R., and Lampen, J. O. Biosynthesis of yeast mannan: Inhibition of synthesis of mannan acceptor by cycloheximide. FEBS Letters (in press, 1970).

Cycloheximide rapidly inhibits the synthesis of mannan by growing yeast cells but has little or no effect on the synthesis of glucan. This suggests that mannan (but not glucan) synthesis requires the addition of sugar units to a polypeptide whose formation is insensitive to cycloheximide. This paper demonstrates that cycloheximide inhibits protein and mannan synthesis to a similar extent. This inhibition results in an accumulation of GDP-mannose (the sugar donor for mannan polymers), but the incorporation of mannan from GDP-mannose into endogenous acceptors by particulate preparation was not affected. It is concluded that upon the addition of cycloheximide formation of the acceptor polypeptide is prevented, but glycosylation of the existing peptides is completed and this mannan is incorporated into the cell wall. Glycosylation now stops for lack of acceptors and GDP-mannose accumulates. These data support the hypothesis that the polypeptide moiety of mannan is synthesized on polyribosomes, according to established pathways of protein synthesis and that the carbohydrate chains are probably added during transport of macromolecules from the polysomes to the site of incorporation into the wall. A similar pathway has been proposed for the secretion of glycoproteins by mammalian cells.

- (2) Tkacz, J. S., Cybulska, E. B., and Lampen, J. O. Specific staining of wall mannan in yeast cells with fluorescein conjugated concanavalin A. J. Bacteriol. (in press, Jan. 1971).

A procedure was developed for coupling fluorescein isothiocyanate to concanavalin A (which specifically combines with a variety of poly-

saccharides) and for the subsequent isolation of the reactive conjugate. This conjugate stains Saccharomyces cerevisiae but not Schizosaccharomyces pombe or Rhodotorula glutinis, two organisms whose cell walls do not contain branched homopolymers of  $\alpha$ -linked mannose. The staining of S. cerevisiae is competitively inhibited by methyl- $\alpha$ -D-mannopyranoside. It is concluded that the staining of S. cerevisiae results from the specific interaction of the fluorescein concanavalin A conjugate with the  $\alpha$ -mannan of the cell wall. This technique is being used to determine the points on the cell at which mannan is inserted during bud formation.

- (3) Ghosh, B. K. Grooves in the plasmalemma of S. cerevisiae, seen in glancing sections of double aldehyde fixed cells. J. Cell Biol. (in press, January 1971).

It has been observed that a double aldehyde prefixation (according to Karnovsky) followed by  $\text{OsO}_4$  fixation is superior to current methods in preserving membrane structures of S. cerevisiae. With this improved technique, the ultrastructure of plasmalemmal invaginations has been clarified. The structures as now visualized by examination of ultrathin sections are consistent with those seen in freeze-etched preparations of similar cells. In stationary phase cells the main feature is 200 to 300 Å wide inpocketings containing fibrillar and globular material which form interconnected grooves on the protoplast surface.

- (4) B. K. Ghosh and S. J. Kraeger. Dynamics of plasmalemmal invagination in the synchronized cells of Saccharomyces cerevisiae. Proc. 28th Annual Meeting, Electron Microscope Society of America, pp. 82-83, 1970.

Stationary phase single cells of S. cerevisiae strain 1829 were separated by discontinuous sorbitol density gradient. When inoculated in 1% yeast extract - 1% glucose medium, these cells were synchronized for 2-3 generations with a generation time of approximately 2 hrs.

Four different kinds of invaginated structures were noted before budding: (1) myelin-like lamellated membranous bodies; they have many 25 Å thick alternate electron dense and lucid layers; these lamellar stacks of membrane always enclose a vesicular space. (ii) folding and refolding of membrane parallel to plasmalemma; (3) rolled up tubules to form vesicles, occasionally these vesicles have concentric layers of tubules; and (4) cluster of vesicles occasionally arranged in 'honeycomb' fashion. The observations suggest that there is extensive membrane growth during the lag period prior to budding. The formation of membranous bodies of diverse organization associated to plasmalemma, might be related to the genesis of cytoplasmic organelles.

- (5) Lampen, J. O. Yeast and Neurospora Invertases. The Enzymes (Paul D. Boyer, ed.), Third Edition (in press, 1971).

CONTENTS OF CYTOCHROMES IN YEAST

Erkki Oura and Heikki Suomalainen

J. Inst. Brewing (In press)

The contents of cytochromes in yeast were determined quantitatively from the absorption spectra, using a solid cell paste of intact yeast. During the industrial production of baker's yeast, the contents of the cytochromes, particularly of cytochrome  $aa_3$  at successive stages, increased gradually with increasing aeration. In semi-aerobically grown baker's yeast, the contents of cytochromes  $aa_3$ , b and c were 0.9, 2.9 and  $2.9 \times 10^{-5}$  moles/litre of fresh yeast (total amount  $6.7 \times 10^{-5}$  moles/litre), while in vigorously aerated commercial baker's yeast the respective values were 2.3, 4.8 and  $5.2 \times 10^{-5}$  moles/litre (total amount  $12.3 \times 10^{-5}$  moles/litre).

In brewer's yeasts separated after the brewing process, the contents of cytochromes were markedly lower than in baker's yeast grown with limited aeration, whereas in top-fermenting yeast the total cytochrome content,  $aa_3 + b + c$ , was in some samples markedly higher,  $7.1 \times 10^{-5}$  moles/litre, than in bottom-fermenting brewer's yeast,  $2.4 \times 10^{-5}$  moles/litre. When brewer's bottom yeast was grown on a laboratory scale under increasing aeration, a maximum appeared in the cytochrome contents when aeration was moderate, and increased aeration inhibited the formation of cytochromes. The cytochrome contents in brewer's bottom yeast may exceed the amounts found in commercial baker's yeast. In addition to aeration, the type of metabolism influences the amounts of cytochromes in yeast.

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YEAST AND ITS EFFECT ON THE FLAVOUR OF ALCOHOLIC BEVERAGES

Heikki Suomalainen

J. Inst. Brewing (In press)

Within the group of more than one hundred identified aroma components, mainly the same substances appear in the volatile aroma fraction of beer, wine, and distilled beverages. In view of this, it seems evident that the raw materials utilized for the production of the beverages, contribute to no more than a limited extent towards the aroma composition. The aroma components which are most noticeable are produced by the yeast during fermentation, and the nature of the final aroma first and foremost depends upon the kind of yeast used, and upon the fermentation conditions. Although the yeast for the most part produces the same aroma components also in different beverages, considerable variances may occur in the quantities of aroma components even in the beverages of the same type.

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$\alpha$ -HYDROXY KETONES, ACETOIN AND HYDROXY PENTANONE, IN WINES

Pentti Ronkainen, Saara Brummer and Heikki Suomalainen

Am. J. Enol. Viticult. (In press)

$\alpha$ -Hydroxy ketones, acetoin and hydroxy pentanone (3-hydroxy-2-pentanone and/or 2-hydroxy-3-pentanone) have been isolated from eight different European white wine brands and eight red wine brands and converted in acid solution to the corresponding vicinal diketones by steam distillation. The diketones formed have been analyzed by head-space gas chromatography of the distillate, with the aid of an electron capture detector. The mean contents of acetoin and hydroxy pentanone were in white wines 16 mg/l and 0.8 mg/l, the respective values in red wines being 46 mg/l and 3 mg/l.

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DIACETYL AND FORMIC ACID AS PARALLEL DECOMPOSITION  
PRODUCTS OF 2-ACETOLACTIC ACID

Pentti Ronkainen, Saara Brummer and Heikki Suomalainen

Acta Chem. Scand. (In press)

In fermentation solutions, 2-acetolactic acid appears as an intermediate in valine synthesis; it is unstable, and decarboxylates readily to acetoin, both enzymatically, and under the influence of strong mineral acids. Acetolactic acid also tends towards spontaneous decomposition, with or without the contribution of certain metal cations, and also in fermentation solutions; one of the products of spontaneous decomposition is diacetyl and another is formic acid. According to the head-space analyses, acetolactic acid decomposes within the range of pH 5-6, giving 40-60 mole per cent of diacetyl as calculated from the starting material. The percentage of corresponding formic acid is often lower than that of diacetyl, by virtue of the method of isolation and analysis applied; in some cases, however, we have found equivalent contents of diacetyl and formic acid.

- XV A. Ü. Ziraat Fakültesi Fermantasyon Teknolojisi Kürsüsü, Ankara, Turkey.  
Communicated by M. Hilmi Pamir.

The following paper will appear in December issue of "Yearbook of the Faculty of Agriculture".

EFFECT OF YEAST STRAINS ON THE FORMATION OF SO<sub>2</sub>-BINDING  
COMPOUNDS AND ON THE SO<sub>2</sub>-BINDING RATE IN WINES

Summary

In this study 7 native yeast strains were tested on SO<sub>2</sub>-binding compounds (acetaldehyde, pyruvic and  $\alpha$ -ketoglutaric acids) under the selected conditions corresponding to those of southern vineyard areas e.g. high temperature and sugar concentration, low acidity. The highest binding effects for most strains were demonstrated at 35C, 90 degrees Oechsle and pH 4.18. For example, the bound SO<sub>2</sub> rate increased from 58.8% at 25C to 90.2% at 35C. This situation

seems to be connected firstly with the high concentrations of pyruvic acid and secondly with  $\alpha$ -ketoglutaric acid at 35C, but not with acetaldehyde, because acetaldehyde decreased in many cases at this temperature. On the other hand, a sugar concentration of 90 degrees Oechsle resulted in an increase during fermentation at 25C up to 95.8% with respect to the bound SO<sub>2</sub> rate, whereas 100 degrees Oechsle retarded the formation of SO<sub>2</sub>-binding mainly according to the amount of pyruvic and  $\alpha$ -ketoglutaric acids. Moreover the H<sup>+</sup>-concentration stimulated obviously and generally the formation of the binding compounds in wine up to 4.18. For this reason the bound SO<sub>2</sub> rate increased up to 89.4%. After this concentration both the concentrations of SO<sub>2</sub>-binding compounds, especially with respect to pyruvic and  $\alpha$ -ketoglutaric acids, and also the rate of SO<sub>2</sub>-binding decreased down to 44.4%. As a result of these assays we can say that the yeasts Nr. 1,3 and 4 were especially selected. If these optimum conditions dominate separately or together during a fermentation, the SO<sub>2</sub> consumption of wine increases accordingly. In this case free SO<sub>2</sub> is not found in practice due to binding of almost all of the SO<sub>2</sub>, even if it is added at ca. 200 mg/l total SO<sub>2</sub> after fermentation. For this reason it is very difficult to protect such a wine by SO<sub>2</sub> addition against biochemical changes. From this point of view the effectiveness of these three factors it was observed that sugar concentration, temperature and pH were significant, respectively, for the formation of SO<sub>2</sub>-binding compounds in wines. Moreover in this study it was also demonstrated, if SO<sub>2</sub> addition to the must before fermentation changes proportionally this binding effect. In this case the consumption of SO<sub>2</sub> was higher in order to maintain a suitable free SO<sub>2</sub> level due to the stimulation of acetaldehyde formation.

XVI The State Institute for Technical Research, Biotechnical Laboratory, Box 12192, Helsinki 12, Finland. Communicated by T. M. Enari.

#### EFFECT OF WORT AMINO ACIDS ON FERMENTATION

M. Linko, M. Loisa, V. Mäkinen ja T.-M. Enari

MBAA Technical Quarterly, in press

The effects of lack or excess of wort amino acids on the fermentation rate, the formation of diacetyl and fusel oil, the growth of yeast and the usage of individual amino acids were investigated.

The wort (prepared of Pirkka high enzyme malt) and sucrose solution were blended to obtain mixtures containing 10 to 100 per cent of wort and 20 to 0 per cent of sucrose solution.

17.3 mg  $\alpha$ -amino nitrogen per litre (30 per cent wort in the mixture) was needed to ensure normal fermentation rate, sufficient growth of yeast and normal metabolic pathways. On the other hand, an excess of amino acids did not cause very much changes as compared with the minimum amount.

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## INFLUENCE OF NITROGENOUS COMPOUNDS ON BEER FERMENTATION

V. Mäkinen, Auli Haikara and T.-M. Enari

Suomen Kemistilehti 43 (1970), n:o 11

The effect of amino acid concentration of wort on fermentation rate, yeast growth, uptake of amino acids by yeast and production of fusel alcohols in aerated stirred fermentation, anaerobic stirred fermentation and unstirred fermentation has been studied.

Two worts, one produced from a low-nitrogen malt and one produced from a high-nitrogen malt, were used. Two yeast strains, one very flocculent and one non-flocculent, were compared.

A higher amino acid concentration of the wort increased the rate of fermentation and yeast growth.

The rates of uptake of different amino acids depended on the concentrations of the amino acids. The rate of absorption decreased when the original concentration of amino acids increased. Some amino acids absorbed slowly or not at all when the wort nitrogen concentration was high. In unstirred fermentation both yeasts formed more fusel alcohols when the wort nitrogen concentration was high. High wort nitrogen caused a decrease in the formation of fusel alcohols when the non-flocculent yeast was used in stirred fermentations but caused a large increase when the flocculent yeast was used.

### XVII Brief News Items

1. The Editor announces with deep regret the untimely death of Professor Augusto Capriotti on April 10, 1970. Professor Capriotti who taught at the University of Sassari, Italy, was killed in an automobile accident. His bibliography of 85 pages has been recorded in "Studi Sessaresi", Sez III, Vol. XVIII, 1-10, 1970.
2. A new catalogue of the National Collection Yeast Cultures has been published by the Brewing Industry Research Foundation, Nutfield, Surrey, England, Director A. H. Cook. Publication date Jan. 1970. The collection is maintained at the B.I.R.F.
3. Division of Biological and Medical Research, Argonne National Laboratory, Argonne, Illinois, 60439. Reported by F. Schlenk and G. Svihla.

Our studies on the penetration of proteins through the yeast cell wall (Candida utilis and Saccharomyces cerevisiae) and their effect on the cell membrane have resulted in the following publications:

Ultraviolet micrography of penetration of exogenous cytochrome c into the yeast cell. G. Svihla, J. L. Dainko, and F. Schlenk. J. Bacteriol. 100, 498-504 (1969).

Enzymatic activity of yeast cell ghosts produced by protein action on the membranes. F. Schlenk and C. R. Zydek-Cwick. Arch. Biochem. Biophys. 138, 220-225 (1970).

The accumulation and intracellular distribution of biological sulfonium compounds in yeast. F. Schlenk, J. L. Dainko, and G. Svihla. Arch. Biochem. Biophys. (September issue, 1970).

Dr. Kathleen A. Killick, formerly of the Illinois Institute of Technology, has completed her sojourn at Argonne. She is continuing her research at the Boston Biomedical Research Institute, Boston, Mass. 02114.

4. Fred Sherman, Department of Radiation Biology and Biophysics, The University of Rochester, School of Medicine and Dentistry, Rochester, New York 14620, writes: Below are cited two recent publications:

Sherman, F., J. W. Stewart, J. H. Parker, G. J. Putterman, B. B. L. Agrawal, and E. Margoliash

The relationship of gene structure and protein structure of iso-1-cytochrome c from yeast.

Soc. Expt. Biol. Symp. No. 24, "The Control of Organelle Development", 1970, pp. 85-107. Edited by P. L. Miller, Cambridge University Press.

5. Contributed by Leslie R. Hedrick, 14225 SW 150th Avenue, Portland, Oregon 97223. (Professor Emeritus from Illinois Institute of Technology).

Two articles resulting from our research upon the types of yeasts associated with freshwater habitats receiving different types of pollution have been published in the recent issue of Antonie van Leeuwenhoek.

L. L. Woollett and L. R. Hedrick (1970) Ecology of yeasts in polluted water. Antonie van Leeuwenhoek 36: 427-435.

L. L. Woollett, L. R. Hedrick and M.-G. Tarver. A statistical evaluation of the ecology of yeasts in polluted water. Antonie van Leeuwenhoek 36: 437-444.

6. University of Rhode Island, Narragansett Marine Laboratory, Graduate School of Oceanography, Kingston, Rhode Island 02881. Mr. Raja Seshadri writes:

"I am a pre-doctoral student interested in yeasts. My present thesis work involves a comprehensive study of yeasts, epiphytic on seaweeds and their role in the biodegradation of organic matter released by seaweeds. I have recently completed an annual survey of yeast distribution on different seaweeds commonly available on the Rhode Island coast. A total of 300 yeast isolates have been examined,



and their distribution, ecology, taxonomy and physiology studied. I plan to explore the biochemical aspects more deeply in the future. Physiological characteristics reveal interesting phenol utilization patterns seasonally and can be related to seaweed exudation chemistry. All information pertaining to yeasts-seaweed relationships will be incorporated in my forthcoming thesis.

Publications wanted on yeasts utilizing phenolic compounds.

Major Professor: Dr. John McN. Sieburth"

7. Bacteriologisch-Serologisch Laboratorium der Rijksuniversiteit, Oostersingel 59, Groningen, Holland.

N. J. W. Kreger-van Rij reports the appearance of the following publication:

N. J. W. Kreger-van Rij and M. Veenhuis, An electron microscope study of the yeast Pityrosporium ovale. Arch. Mikrobiol. 71: 123, 1970.

Accepted for publication:

N. J. W. Kreger-van Rij and M. Veenhuis, Septal pores in Trichosporon cutaneum. Sabouraudia.

N. J. W. Kreger-van Rij and M. Veenhuis. Bipolar budding in yeasts - an electron microscope study. Antonie van Leeuwenhoek.

8. Ecole Nationale Supérieure Agronomique de Montpellier, Chaire de Génétique, Montpellier, France.

Professor Pierre Galzy reports on the following publications:

- On a modification of the cell wall of certain "smooth colonie" mutant strains of Saccharomyces cerevisiae HANSEN.  
C. BIZEAU, P. GALZY, J. M. BASTIDE et Madeleine BASTIDE.  
Abstract - The presence of the genes  $pl_1$ ,  $pl_5$  or  $pl_7$  provokes a modification of the cell wall. It is possible that each controls one step of the synthesis of the same compound of the cell wall.  
Prague - 1st International Symposium Genetics of Industrial Microorganisms August 23-28, 1970.
- Etude de la préparation de protoplastes chez 2 souches de Saccharomyces cerevisiae HANSEN.  
C. BIZEAU, et Emilienne BIZEAU.  
The study of the preparation of protoplasts of two yeast strains shows that certain "smooth colonie" mutants are more sensitive to the Helix enzyme action.  
Annales de Technologie - in press.
- Genetic marking of industrial yeasts.  
Monique RADIER, P. GALZY et J. PASERO\*  
Laboratoire de Recherches de la Chaire de Génétique  
INRA - ENSA - Montpellier, France.  
\*Division de Microbiologie S. F. B. P. Lavera, France.  
This paper gives the results of a systematic study with the aim of

marking industrial strains. Mutants resistant to various ions ( $\text{Cd}^{++}$ ,  $\text{Co}^{++}$ ,  $\text{Cu}^{++}$  or  $\text{AsO}_4^{---}$ ) have been isolated and studied. Prague - 1st International Symposium Genetics of Industrial Microorganisms August 23-28, 1970.

9. Professor A. Castellani, Escola Nacional de Saude Publica e de Medicina Tropical, Lisbon, Portugal, reports the publication of the following two publications:

Tinea Nigra Unguim, A. Castellani, Journal of Tropical Medicine and Hygiene, Vol. 71, pages 74 and 75, 1968.

A Note on Infantile and Juvenile Leucodermata and Their Palliative Cosmetic Treatment, A. Castellani, Journal of Tropical Medicine and Hygiene, Vol. 73, pages 208-209, 1970.

10. Dr. William A. Clark, Director of the American Type Culture Collection, 12301 Parklawn Drive, Rockville, Maryland 20852, writes:

The ATCC is interested in having yeast workers who describe new species deposit type cultures in the American Type Culture Collection as well as in other collections. We feel that duplication of these important cultures in several collections is important.

Doctor Sally Meyer will join our staff in 1972 and she will be responsible for our yeast collection.

11. Instituto Nacional de Investigaciones Agronomicas, Avda. de Puerta de Hierra, Madrid 3, Spain. Prof. J. Santa Maria has sent the following recent publications to the editor:

J. Santa Maria y D. Vidal, Segregación anormal del "mating tipe" en Saccharomyces. Boletín número 62 del Instituto Nacional de Investigaciones Agronómicas, Junio 1970.

J. Santa Maria, Saccharomyces gaditensis y Saccharomyces cordubensis, dos nuevas especies de levaduras de "flor". Boletín número 62 del Instituto Nacional de Investigaciones Agronómicas, Junio 1970.

J. Santa Maria y Consuelo Sanchez, Significación taxonómica de las propiedades fisiológicas de las especies incluidas en el género "Kluyveromyces". Boletín número 62 del Instituto Nacional de Investigaciones Agronómicas, Junio 1970.

J. Santa Maria y Concepcion Garcia Aser, Pichia castillae, nov. spec., aislada de insectos. Boletín número 62 del Instituto Nacional de Investigaciones Agronómicas, Junio 1970.

12. The new edition of "The Yeasts - a taxonomic treatise", J. Lodder editor, has now been published (fall 1970). The table of contents and the authors follow below.

Publisher: North Holland Publishing Company, Amsterdam, 1970.

I. General Classification of the Yeasts, J. Lodder

- II. Criteria and Methods Used in Classification, J. P. van der Walt
  - III. Introduction to the Chapters IV, V, VI and VII, and Key to the Genera, J. Lodder
  - IV. Discussion of the Genera Belonging to the Ascomycetous Yeasts, L. J. Wickerham, N. J. W. Kreger-van Rij, J. P. van der Walt, H. J. Phaff, W. Ch. Slooff, M. W. Miller, N. van Uden and L. do Carmo-Sousa.
  - V. Discussion of the Yeast-like Genera Belonging to the Ustilaginales, J. W. Fell, H. J. Phaff and S. Y. Newell.
  - VI. Discussion of the Yeast-like Genera Belonging to the Sporobolomycetaceae, H. J. Phaff.
  - VII. Discussion of the Genera of Asporogenous Yeasts not Belonging to the Sporobolomycetaceae, J. P. van der Walt, N. van Uden, H. Buckley, H. J. Phaff, J. W. Fell, L. do Carmo-Sousa, W. Ch. Slooff, D. G. Ahearn, N. J. W. Kreger-van Rij and M. Vidal-Leiria.
13. Donald G. Ahearn, Department of Biology, Georgia State University, Atlanta, Georgia 30303 writes: The proceedings of the symposium "Recent Trends in Yeast Research", held at Plattsburgh, New York on August 15-16, 1969 has been published and distributed to Libraries and Authors. The publication is a special edition of Spectrum, Monograph Series in the Arts and Sciences published by Georgia State University. Reprints of all contributions will be available from the authors. The contents of the proceedings are listed below.
- I. Taxonomic Significance of the DNA Base Composition in Yeasts. Sally A. Meyer and H. J. Phaff.
  - II. Correlation of Deoxyribonucleic Acid Base Percentages and Taxonomic Characteristics in Hansenula. Lynferd J. Wickerham.
  - III. Yeasts with Heterobasidiomycetous Life Cycles. Jack W. Fell.
  - IV. Clamp Connections in Two Strains of Cryptococcus neoformans. Jean Shadomy.
  - V. Some Recent Observations on Spore Formation and Germination in Saccharomyces. J. J. Miller.
  - VI. Sexuality in Candida lipolytica. Lynferd J. Wickerham, Cletus P. Kurtzman, and Alberta I. Herman.
  - VII. Amine Utilization by Yeasts. Samuel P. Meyers and Mary Ellen Nicholson.
  - VIII. Effects of Pollution on the Seasonal Population of Yeasts in Lake Champlain. W. L. Cook.
  - IX. Ecology and Physiology of Yeasts of an Asphalt Refinery and Its Watershed. W. E. Turner and D. G. Ahearn.

- X. Growth of Yeasts on Hydrocarbons. Vladimir Munk.
- XI. Nutritional Studies on Riboflavin Overproduction by Ashbya gossypii. Louis Kaplan and Arnold L. Demain.
- XII. Fluorescent Antibody Studies of the Black Yeasts. Dorene L. Setliff and C. J. K. Wang.
- XIII. The Synthesis of Starch-like Compounds by Cryptococcus laurentii. Mohamed Salah Foda and Herman J. Phaff.
- XIV. Inhibition of Respiration in Baker's Yeast (Saccharomyces cerevisiae) by 2,4-dinitrophenol. Daniel C. Lee.

W. L. Cook, formerly of State University College of Arts and Science, Plattsburgh, New York, has joined the staff of our Microbiology Division of the Department of Biology. Recent publication on yeasts from the Division:

Standard, P. G. and D. G. Ahearn. 1970. Effects of alkylbenzene sulfonates on yeast. *Appl. Microbiol.* 20: 646-648.

#### XVIII International Meetings and Events.

The following letter was sent by Dr. S. M. Martin to Dr. H. Iizuka:

Japanese Federation of Culture Collections,  
c/o Prof. H. Iizuka,  
Institute of Applied Microbiology,  
University of Tokyo,  
Tokyo, Japan.

Dear Sirs:

At its meeting in Mexico City, 9 August 1970, the Section on Culture Collections, I.A.M.S., voted to dissolve and to be reconstituted as the World Federation for Culture Collections. With this action the resolution passed at the Tokyo Conference on Culture Collections was implemented.

As Chairman of the newly established W.F.C.C. I would like to express, on behalf of our organization, our sincerest thanks to the Japanese Federation of Culture Collections for its generous financial support. Without this support the task of setting up the Federation would have been much more difficult.

Yours sincerely,

S. M. Martin, Chairman,  
World Federation for  
Culture Collections.

SECTION ON CULTURE COLLECTIONS

Report to the General Assembly, I.A.M.S., Mexico City, 14 Aug. 1970

I wish to report that The Section on Culture Collections has voted to dissolve and to be reconstituted as the World Federation for Culture Collections (WFCC). The statutes of the WFCC, as amended, have been accepted at an open meeting, pending acceptance of I.A.M.S. The elected officers of the WFCC are:

Chairman -	Dr. S. Martin (Canada)
Vice-chairman -	Prof. H. Iizuka (Japan)
Secretary -	Dr. S. Lapage (U.K.)
Treasurer -	Prof. V. Skerman (Australia)

As Chairman of the newly formed WFCC, I would like to express our heartfelt thanks to the Japanese Federation of Culture Collections for its generous financial support in setting up this federation.

A most successful Conference on Culture Collections was held in Tokyo, in 1968, under the chairmanship of Prof. Iizuka. The conference proceedings have been published recently by the University of Tokyo Press. The newly formed WFCC has endorsed the recommendation of the Tokyo conference that the next conference on culture collections be held in Brno, Czechoslovakia in 1972 subject, of course, to final acceptance by Czechoslovakia.

The World Survey of Culture Collections, sponsored by the Unesco/ICRO Panel for Microbiology, has proceeded smoothly. It is our hope that the WFCC will be able to publish the World Directory and List of Species not later than June 1971. At about the same time the WFCC will be able to function efficiently as a world data center. For the time being, the focal point for this center will be at the University of Queensland, Brisbane, Australia, and will be under the direction of Prof. Skerman.

The WFCC intends to go ahead with the preparation of an International Handbook of Procedures, in conjunction with the Sub-committee on Numerical Taxonomy, I.C.N.B.

The WFCC is attempting to establish a Fellowship and Training Course program, in conjunction with the Unesco/ICRO Panel for Microbiology, as an aid to microbiology in developing countries.

The WFCC is trying to implement the recommendations of the Tokyo conference relating to the establishment of a network of centers for the identification of microorganisms.

Finally, the WFCC wishes to extend a vote of thanks to Prof. Skerman for his untiring work as President of the Section on Culture Collections.

S. M. Martin