APPLIED CHEMISTRY SECTION

FERMENTATION INDUSTRIES DIVISION
COMMISSION ON CHARACTERIZATION AND EVALUATION OF DRIED YEAST*

SURVEY OF WORK DURING THE YEARS 1957-1962

The work has concerned the creation of standards for inactive dry yeast and development of reference methods of analysis.

Collaborative analyses have been performed by different laboratories on two different samples of dry yeast. The first sample was sent by Klokgieters (Koninklijke Nederlandsche Gist- en Spiritusfabriek N.V., Delft) and the following laboratories collaborated in the analyses:

Institut für Gärungsgewerbe und Stärkefabrikation, Berlin, Germany
Zellstoffabrik Waldhof, Mannheim-Waldhof, Germany
Koninklijke Nederlandsche Gist- en Spiritusfabriek N.V., Delft, Holland
Fleischmann Laboratories, Stamford, Connecticut, U.S.A.
National Yeast Corporation, Belleville, N.J., U.S.A.
Tate & Lyle Ltd., Keston, Kent, England
Glaxo Laboratories Ltd., Greenford, England
Laboratoire de l'Institut Professionel de Controle et de Recherches Scientifiques de l'Alimentation Animale, Paris, France
Laboratoire d'étude et de Controle de la qualité des Blés Francais, Paris, France
Service de Chimie Biologique, Faculté des Sciences, Montpellier, France
Laboratoire de Recherches du Service de Santé de la F.O.M., Marseille, France
Laboratoire Municipal de Bordeaux, France
Laboratoire de l'Union Nationale des Groupements de Distillateurs d'Alcool, Paris, France
Laboratoire de Biochimie de la Nutrition, Paris, France
Laboratoire de Physiologie de la Faculté de Pharmacie, Paris, France
Institut Pasteur, Paris, France
Royal Danish Technical University, Copenhagen, Denmark
Versuchsstation für das Gärungsgewerbe, Vienna, Austria.

The second sample was delivered by Mr R. F. Light (Standard Brands Inc., New York, U.S.A.) and the following laboratories have collaborated in the analysis:

Anheuser Busch Inc., St. Louis, U.S.A., Dr Robert Seeley

* R. F. Light, President (U.S.A.), C. N. Frey, Vice-President (U.S.A.), H. Suomalainen, Secretary (Finland), P. Birolaud (France), G. Butschek (Germany), J. M. Klokgieters, Secretary 1958–1961 (Netherlands), W. F. J. Cuthbertson (U.K.), H. Jørgensen (Denmark), H. Lundin, President 1957–1961 (Sweden), F. Parisi (Italy), A. Szilvinyi (Austria).

Department of Health, Education and Welfare, Washington, D.C., U.S.A., Dr Daniel Banes
Department of National Health and Welfare, Ottawa, Ontario, Canada, Dr J. A. Campbell
The Fleischmann Laboratories, Stamford, Connecticut, U.S.A., Mr W. E. Maynard
Institut für Gärungsgewerbe und Stärkefabrikation, Berlin, Germany, Dr A.-M. Herbst
Koninklijke Nederlandsche Gist- en Spiritusfabriek N.V., Delft, Holland, Mr J. M. Klokgieters
National Yeast Corporation, Belleville, N.J., U.S.A., Dr Morris Mead
Red Star Yeast, Milwaukee, Wisconsin, U.S.A., Dr Gerald Reed
Research Laboratories of the State Alcohol Monopoly (Alko), Helsinki, Finland, Dr Heikki Suomalainen
Statens Institut för Folkhälsan, Stockholm, Sweden, Dr Vidar Hellström
Statens Vitaminlaboratorium, Copenhagen, Denmark, Prof. Willy Hjarde
Syndicat des Producteurs de Levure-Aliment de France, Paris, France, Mr Pierre Birolaud
The Upjohn Company, Kalamazoo, Michigan, U.S.A., Mr G. V. Slagel
Vereinigte Mautner Markhof'sche Presshefe Fabriken, Vienna, Austria, Prof. A. Szilvinyi
Vitaminlaboratoriet, Bergen, Norway, Dr Olaf R. Braekkan
Zellstoffabrik Waldhof, Mannheim-Waldhof, Germany
Université de Louvain, Institut Agronomique, Section de Brasserie, Louvain, Belgium, Prof. Joseph Frateur

A preliminary outline of the standards and reference methods was made by Klokgieters and they have been finally formulated at the Meeting of the Commission B of the Fermentation Industries Division, September 16th, 1962, London. The survey of results of the final analyses and the statistical summary were made at the Research Laboratories of the State Alcohol Monopoly (Alko), Helsinki, Finland.

DEFINITION OF DRIED YEAST ACCORDING TO I.U.P.A.C. SPECIFICATION

By " dried yeast "* is meant the dried whole organism of one individual yeast or of a mixture of several yeasts belonging to the family Saccharomycetaceae, sub-family Saccharomycetoideae, and to the family Cryptococcaceae, sub-family Cryptococcoideae and Rhodotoruloideae (classification of Lodder and Kreger-van Rij), obtained either as a by-product of fermentation processes or by special culture and conforming to such standards as may be laid down.

So-called " fat yeasts " (for instance *Cryptococcus terricolus*, *Rhodotorula* species *etc.*) containing more than 20 per cent fat are not considered here.

* In France this should be understood as " levure-aliment sèche " and not " levure sèche ", as the latter term may also designate an active dry yeast for bread making, wine making, *etc.*

It is desirable that the origin or origins of the yeast be specified by the manufacturer and the strain of yeast be identified. The substrate on which the yeast is grown shall be stated.

It is not permitted to sell under the label " Dried Yeast " yeasts which have been extracted or which contain inert fillers or substances that are not incorporated components of normal yeast cells .

Any additions to yeast, even of those nutrients normally present in yeast should be declared.

It is emphasized that a dried yeast sample may still be satisfactory for a number of purposes even though it may not meet the I.U.P.A.C. standards.

STANDARDS OF DRIED YEAST ACCORDING TO I.U.P.A.C. SPECIFICATIONS

1. **Moisture**
   Maximum 10 per cent

2. **Total nitrogen in air-dried yeast**
   7·2 per cent and over

3. **Ash in air-dried yeast**
   Maximum 10 per cent

4. **Lead in air-dried yeast**
   Maximum 5 mg/kg

5. **Arsenic in air-dried yeast**
   Maximum 5 mg/kg

6. **Vitamins in air-dried yeast**

   Thiamine
   Not less than 10 mg/kg

   Riboflavin
   Not less than 30 mg/kg

   Niacin
   Not less than 300 mg/kg

7. **Starch in air-dried yeast**
   No blue colour detectable in the iodine test

8. **Bacteria and Mold count in air-dried yeast**
   The live bacteria count not over 7500 per g and the mold count not over 50 per g

9. **Salmonella in air-dried yeast**
   Absent

Table 1. Survey of the results obtained in the collaborative

		Moisture (%)	Total nitrogen % in original substance	Phosphorus (% o.s.)	Fat (% o.s.)	Ash (% o.s.)	Lead (mg/kg o.s.)	Iron (mg/kg o.s.)	Arsenic (mg/kg o.s.)	Thiamine (mg/kg o.s.)
(1)	Anheuser Busch Inc., *St. Louis*	5·03	8·2	1·28	2·7	5·5	—	35	—	137·5
(2)	Department of Health, Education and Welfare, *Washington*, D.C.	5·17	7·85	—	—	5·27	0·5	—	0·5	134
(3)	Department of National Health and Welfare, *Ottawa*	—	—	—	—	—	—	—	—	148
(4)	The Fleischmann Laboratories, *Stamford*	5·23	7·86	1·32	—	5·30	—	34·5	—	—
(5)	Institut für Gärungsgewerbe und Stärkefabrikation, *Berlin*	5·10	7·93	1·29	3·59	5·22	2·6	38·2	0·49	128
(6)	Koninklijke Nederlandsche Gist- en Spiritusfabriek N.V., *Delft*	4·91	7·82	1·23	—	5·55	—	28	—	143
(7)	National Yeast Corporation, *Belleville*	5·08	7·83	—	—	5·20	—	—	—	—
(8)	Red Star Yeast, *Milwaukee*	5·16	7·87	—	—	—	—	—	—	127
(9)	Research Laboratories of the State Alcohol Monopoly, *Helsinki*	5·32	7·52	1·31	2·62	4·90	1·5	26·1	0·38	132
(10)	Statens Institut för Folkhälsan, *Stockholm*	—	—	—	—	—	—	—	—	129
(11)	Statens Vitamin-Laboratorium, *Copenhagen*	4·78	7·9	1·31	—	5·25	—	37·5	—	122
(12)	Syndicat des Producteurs de Levure-Aliment de France, *Paris*	4·7[b] 4·4[f]	7·73[b] 7·84[f]	—	—	5·25[b] 5·3[f]	Traces[b]	—	—	119[c]
(13)	The Upjohn Company, *Kalamazoo*	4·49	7·95	1·4	—	5·2	<5	40	<3·5	129
(14)	Vereinigte Mautner Markhof'sche Presshefe Fabriken, *Vienna*	5·80	7·67	—	—	5·16	—	—	—	—
(15)	Vitaminlaboratoriet, *Bergen*	4·90	—	—	—	—	—	—	—	135
(16)	Zellstoffabrik Waldhof, *Mannheim-Waldhof*	5·02	7·92	—	—	5·01	0·90	—	0·48	—
(17)	Université de Louvain, Institut Agronomique, Section de Brasserie, *Louvain*	5·05[g]	7·80[g]	—	—	5·25[g]	0·35[g]	—	0·05[g]	119[h]

[a] Performed by Staatliche Medizinal-Untersuchungsamt, Hannover.
[b] Performed by Laboratoire de l'Union Nationale des Groupments de Distillateurs d'Alcool.
[c] Performed by Faculté de Pharmacie de Paris, Laboratoire de Physiologie.
[d] Performed by Laboratoire de Bacteriologie Alimentaire.

analysis of the I.U.P.A.C. Dry Yeast Sample

Riboflavin (mg/kg o.s.)		Niacin (mg/kg o.s.)		Pyridoxin (mg/kg o.s.)	Pantothenic acid (mg/kg o.s.)		Crude fibre	Starch	Bacteria and mold (count/g o.s.)		Salmonella (count/g o.s.)	
Microb	Chem.	Microb.	Chem.		Total	Free			Bact.	Mold		
—	77·6	540	—	—	85·8	39·9	—	Neg.	2750		Neg.	(1)
—	79	568	556	—	—	—	—	Neg.	55	None	Neg.	(2)
—	82·7	—	580	—	—	—	—	—	—	—	—	(3)
—	—	—	—	—	—	—	—	Neg.	—	—	—	(4)
78	—	600	—	6·4	105	36	—	Neg.	300	—	Neg.[a]	(5)
—	72	—	570	—	—	—	—	Neg.	250	<10	—	(6)
—	—	—	—	—	—	—	—	—	—	—	—	(7)
—	94	594	—	—	—	—	—	—	10	<10	Neg.	(8)
77	86	618	—	18·9	103	26·2	—	—	3700	—	—	(9)
81	—	630	—	13·6	—	—	—	—	—	—	—	(10)
73	81	617	460	19	100	36·5	—	—	—	—	—	(11)
—	—	594[c]	—	—	—	—	0·8[b]	Neg.[b]	7200[d] 4000[e]	10[d] None[e]	Neg.[d e]	(12)
78	—	655	—	—	—	—	—	Neg.	<10	20	Neg.	(13)
—	—	—	—	—	—	—	—	—	—	—	—	(14)
65·3	—	649	—	15·5	110·7	—	—	—	—	—	—	(15)
84·9	—	645·9	—	—	95·9	—	—	—	1650	None	Neg.	(16)
—	39·1[h]	—	825[h]	—	—	—	—	—	—	—	—	(17)

[e] Performed by Institut Pasteur, Lille.
[f] Performed by Institut National de la Recherche Agronomique.
[g] Performed by Laboratoire Central du Ministère des Affaires Economiques et de l'Energie.
[h] Performed by Laboratoire de l'Etat (Ministère de l'Agriculture) à Anvers.

REFERENCE METHODS OF ANALYSIS

1. Moisture

The official method of A. L. Brandon (*J. Assoc. Offic. Agr. Chemists* **44**, 394, 1961) for determination of moisture in dry yeast (Method 2: Air oven method, p. 396).

2. Total nitrogen

The official methods of A. L. Brandon (*J. Assoc. Offic. Agr. Chemists* **44**, 394, 1961) for determination of total nitrogen in dry yeast (macro method p. 396 and micro method p. 399). Of the macro methods method 1 is recommended and of the micro methods the method 2; both these use boric acid in the receiver.

3. Ash

Incineration at 590–650 °C, not over 650 ° for 16–20 h in a muffle furnace (Nordisk Metodikk-Komite for Naeringsmidler No. 7, 1952).

4. Lead

The dithizon method described in *A.O.A.C.* (9th ed., p. 317).

5. Arsenic

The generally used Gutzeit method (*A.O.A.C.*, 9th ed., p. 305).

6. Vitamins

Thiamine

The fluorometric thiochrome method of *A.O.A.C.* (9th ed., p. 655); directions for the conversion of disphosphothiamine to thiamine are given.

Riboflavin

The microbiological determination with *Lactobacillus casei* ATCC 7469 as test organism (*A.O.A.C.*, 9th ed., p. 669) and the fluorometric method of Saletan *et al.* (*A.O.A.C.*, 9th ed., p. 658).

Niacin

The microbiological method with *Lactobacillus arabinosus* ATCC 8014 as test organism (*A.O.A.C.*, 9th ed., p. 667).

7. Starch

No detectable amount of starch should be noted in the iodine test, *i.e.* no blue colour should be detectable.

8. Bacteria and Mold count

Determination according to *U.S. Pharmacopeia* (XV, p. 790, 1955; determination not included in the following edition XVI, 1960).

9. Salmonella

Determination according to the method for the examination of brewer's yeast for *Salmonella* supplied by Dr Glenn G. Slocum, Director Division of Microbiology, Bureau of Biological and Physical Sciences, Department of Health, Education, and Welfare, Food and Drug Administration, Washington, D.C.

OTHER RECOMMENDED METHODS

Phosphorus

The method described in Nordisk Metodik-Komite for Levnedsmidler 1954, No. 16.

Iron

The *A.O.A.C.*-method (9th ed., p. 159) for enriched and phosphated flour with *o*-phenanthroline or α,α-dipyridyl, involving wet digestion.

Pantothenic acid

The method for microbiological determination of pantothenic acid in dried yeast as described by H. Klaushofer (*Mitt. Versuchsstat. Gaerungsgewerbe Inst. Angew. Mikrobiol.* **15**, 71, 1961).

Table 2. Statistical calculation of the results obtained in the collaborative analysis of the I.U.P.A.C. Dry Yeast Sample

Analysis	*Number of laboratories* (n)	*Standard deviation* (s)	*Confidence Intervals for the single value* $\bar{X} \pm t_{0.05} \times s$	*Mean of the series* $\bar{X}$	*The limits for the mean* $t_{0.05} \times s/\sqrt{n}$
Moisture (%)	16	0·33	4·31– 5·71	5·01	±0·18
Total nitrogen (% o.s.)	15	0·15	7·54– 8·16	7·85	±0·08
Phosphorus (% o.s.)	7	0·05	1·19– 1·43	1·31	±0·05
Fat (% o.s.)	3	0·54	0·7 – 5·3	3·0	±1·3
Ash (% o.s.)	14	0·16	4·89– 5·59	5·24	±0·10
Lead (mg/kg o.s.)	5	0·92	0– 3·71	1·17	±1·14
Iron (mg/kg o.s.)	7	5·25	21·3 –47·1	34·2	±4·9
Arsenic (mg/kg o.s.)	5	0·19	0– 0·91	0·38	±0·24
Thiamine (mg/kg o.s.)	13	8·7	112–150	131	±5·3
Riboflavin (mg/kg o.s.)					
Microb.	7	6·2	62– 92	77	± 6
Chem.	8	16·4	38–115	76	± 14
Niacin (mg/kg o.s.)					
Microb.	11	35·5	531–689	610	± 24
Chem.	5	135	221–975	598	±169
Pyridoxin (mg/kg o.s.)	5	5·2	0– 29	14·7	±6·4
Pantothenic acid (mg/kg o.s.)					
Total	6	8·6	78–122	100	±9·0
Free	4	5·9	16– 54	35	±9·4

$t_{0.05}$ is a statistical constant for the risk of 5%.