Pilot Extraction Studies. Evaluation of Defatted Yeast and Yeast Fat

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THE purpose in making pilot extraction studies on yeast was primarily to evaluate defatted yeast from a flavor standpoint, and secondarily to evaluate the fat. It is desirable for certain food and pharmaceutical purposes to have a bland tasting yeast.

Preliminary taste tests comparing one gram of extracted yeast in 100 cc. of canned tomato juice with the same concentration of unextracted yeast indicated that the extracted yeast had a blander taste. If special uses could be found for the fat from yeast then it might be advisable to investigate more thoroughly the extraction of fat from a high fat yeast. However, for purposes of obtaining an essentially fat-free yeast from a taste standpoint, the problem was efficiently to extract very small percentages of fat and to reduce the fat content of the marc to as low a concentration as possible. Furthermore, if extensive organoleptic tests showed that consumer preference would be for the fat extracted yeast, then it would be necessary to develop a commercial process for this extraction.

Laboratory analysis by Soxhlet extraction with Skellysolve B for six hours has shown that the crude lipides content of primary grown yeasts normally will vary between .4 and 2%. Several sources (1, 2) have reported however that certain yeasts have a lipides content as high as 60%.

Experimental

A dried, non-viable primary grown yeast, Saccharomyces cerevisiae, in both "chipped," a coarse form, and "powdered," a finely pulverized form, was used for extraction studies.

TABLE I

Screen Analysis of Powdered Yeast

U. S. Standard Mesh No.	Percentage Retained
60	1-2
100	12-13
140	18-20
200	20-22
270	22-24
325	0-2
—325	20-23

It was realized even before preliminary laboratory extraction runs that the chipped and powdered yeast presented different problems. Presumably the extraction efficiency with powdered yeast would be higher than for chipped yeast because of the greater surface area. However, because of the extreme fineness of powdered yeast, as shown in Table I, it was considered a possibility that a good proportion of the fines would end up in the miscella and cause a filtration problem.

The proper solvent was determined by laboratory Soxhlet extractions of small batches of primary grown yeast, using various solvents. Among solvents tried were carbon tetrachloride, benzene, diethyl ether, petroleum ether (30-60°C. boiling range), and Skellysolve B. The alcohols were ruled out as a solvent because of the known solubility of the B-complex vitamins in alcohols. In all cases the extraction was for six hours, and the solvent was removed from the extracted yeast by drying in a vacuum oven at 60°C. for 16 to 20 hours. Because it was difficult to remove the last traces of carbon tetrachloride, benzene, or diethyl ether even at temperatures up to 75°C. in the vacuum oven, these solvents were rejected for further use. No taste difference could be detected between the yeasts extracted with petroleum ether and Skellysolve B, but Skellysolve B was employed for subsequent work because of its wide use in the vegetable oil industry.

Two extraction systems were chosen for investigation: a horizontal and a vertical extractor system were tried on a pilot scale. These systems both embodied the same principle of continuous counter-current extraction; that is, the fresh solvent was in contact with the extracted yeast and the concentrated miscella was in contact with the unextracted yeast. In the case of the horizontal extractor the yeast was conveyed from section to section by impeller blades, whereas the yeast dropped by gravity from plate to plate in the vertical extractor. The recovery of solvent from the miscella was approximately the same in both cases: filtration of the solvent to remove any fines, followed by concentration in a single effect vacuum evaporator to about 95%. The remaining solvent was removed in a bubble cap distillation column under about 20-22" Hg vacuum, using sparging steam. The method of removing the solvent from the marc was different. In the case of the horizontal extractor system the extracted yeast was dried in batches in an air oven at 212°F. and heated by steam coils for 45 minutes, whereas the vertical extractor system used a continuous three-pass steam heated tunnel dryer, employing 15 to 20 p.s.i.g. steam on all three passes. In both cases some oil was lost in the stripper because of the high viscosity of the oil.

Discussion

Results of Horizontal Extractor Studies. Three runs were made in the horizontal extractor (3); the first was on powdered yeast and was a preliminary run to determine the proper extraction conditions; the second, for 33 hours, was to extract the bulk of the powdered yeast; the third run was on chipped yeast. Data for runs 1, 2, and 3 are given in Table II.

A comparison of the data for Runs 1 and 2 shows that increasing the solvent ratio from 1.8 to 2.1 lowered the residual fat in the yeast from .057% to .035% and increased the miscella concentration from 1.2% to 1.8%. In both cases less than 5% of the original yeast ended up in the miscella, and no difficulties were encountered with filtration. On Run 3 (chipped yeast), because the apparent density was much less, the rate of feed was considerably reduced. The residual fat content of the extracted chipped yeast was .084% while the miscella concentration was only .73%; the percentage of feed in the miscella however was only .2%. These data indicate that the powdered yeast is more efficiently extracted than the

TABLE II

Horizontal Extractor Data

Solvent Used Material	Run 1 Skelly-solve B Powdered Yeast	Run 2 Skelly-solve B Powdered Yeast	Run 3 Skelly-solve B Chipped Yeast
Contact Time (min.)	60	60	60
Total Time of Run (hrs.)	9	33	6
Meal Feed Rate (#/hr.)	7.6	7.1	4.1
Density of Feed Meal (#/ft.³)	36.7	36.7	20.4
Oil in Feed Meal (% dry basis)	1.8	1.8	.44
H_2O in Feed Meal (%)	5.2	5.2	5.1
Solvent Rate (#/hr.)	14.0	14.6	9.3
Solvent Ratio	1.8	2.1	2.3
Solvent Feed Temp. (°F.)	73	69-94	72
Extractor Temp. (°F.)	130	129	130
Oil in Extracted Meal (% dry basis)	.057	.035	.084
Oil in Miscella (%)	1.2	1.8	.73
Feed in Miscella (% of feed)	4.2	4.4	.20
Calculated Oil Recovery (%)	96.8	98.2	95.3

chipped yeast and that the fines gave less trouble than anticipated with powdered yeast.

Figure 1 is a semi-log graph of the residual oil content in the marc plotted against time and of the oil content in the miscella plotted against time for

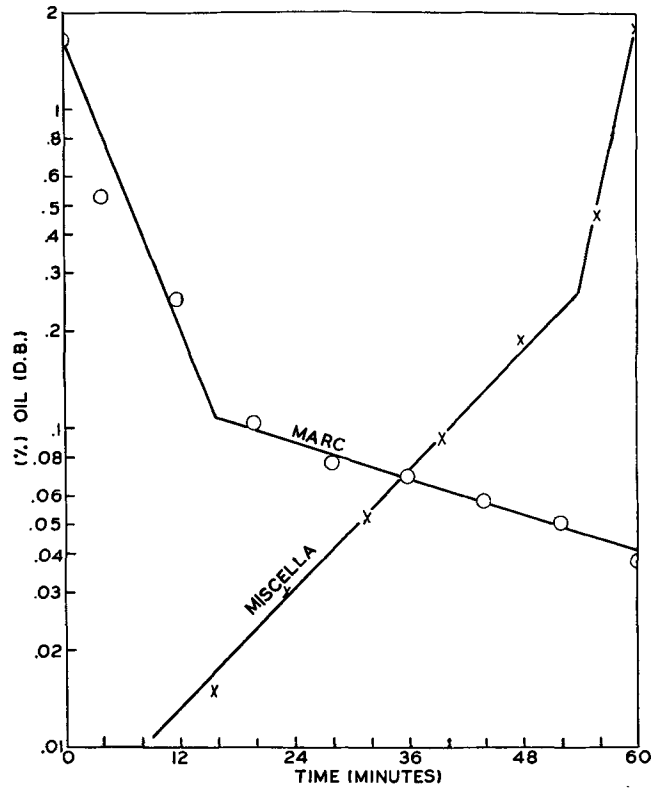

FIG. 1. Marc and miscella concentrations versus time.

Run 2 (powdered yeast). The marc curve is represented approximately by two straight lines of the form:

$$\text{Log } R = m\,t + b$$

where R = % oil remaining in the marc
t = time in minutes
m = logarithmic slope of line
b = logarithm of ordinate intercept
(m and b are constants)

Under the conditions for this run the curve is not quite a straight line above approximately .1% residual oil, but for the nearest straight line through these points the constants m and b are —.087 and .243, respectively; below .1% the constants are —.0085 and —.840, respectively.

Although the break in the curve was about .1% residual oil for Run 2 on powdered yeast, the break occurred at about .07% for chipped yeast. The sharp break in the curve at this point would indicate two separate diffusion processes.

The equilibrium concentration of the miscella corresponding to a marc concentration (R) at a time (t) can be found from the miscella curve.

It is interesting to note from Table IV that, where the residual fat in the marc was .033%, the phosphatides were 47.1% whereas when the residual fat was .45% (i.e., above the break in the curve), the phosphatides were only 18.4%. The likelihood that, below .1%, primarily phosphatides are being extracted can only partially explain this difference.

Results of Vertical Extraction Studies. The vertical extractor studies were conducted primarily for the purpose of obtaining a sufficient quantity of yeast for commercial evaluation. This pilot plant had about five times the capacity of the horizontal extractor pilot plant. Since the horizontal extractor is recognized for its ease in handling extremely fine materials (3) and almost 5% of the powdered yeast ended in the miscella, chipped yeast was used for the vertical extractor runs. As a result, no filtration problem was encountered, with only .4 to .5% of the yeast ending in the miscella. This figure could very likely be reduced still further with an adequate fines settling chamber, which was missing from this particular pilot extractor. Table III gives pertinent data for the vertical extractor.

TABLE III

Vertical Extractor Data

	Run 1	Run 2	Run 3
Solvent Used	n-Hexane	n-Hexane	n-Hexane
Feed to Extractor (#/hr.)	40-45	30	40-45
Solvent Ratio	1.9	1.9	1.9
Solvent Inlet Temp. (°F.)	150	150	150
Extractor Top Temp. (°F.)	120	135-145	135-140
Press. on Top Drier, p.s.i.g. steam	20-15	15	15
Press. on Middle Drier, p.s.i.g. steam	20-15	15	15
Press. on Bottom Drier, p.s.i.g. steam	20-15	15	15
Temp. Vapor Scrubber Water	180	180	180
Press. on Evap. Jacket, p.s.i.g. steam	10-20	20	20
Evaporator, Vapor Outlet (°F.)	200	170	160
Evaporator Inlet (°F.)	145	143	136-142
Press. on Filter (p.s.i.g.)	0-16	20-0	2-17-0
Contact Time (min.)	40	60	40
Stripping Column Vacuum (inches Hg)	22	22	22
Feed Ending in Fines (% of feed)	.4-.5	.4-.5	.4-.5
Average Residual Oil in Marc (%)	.58	.49	.45
Average Oil in Feed (%)	1.01	1.01	1.01
Recovery (%)	42.6	50.5	54.5

When it was learned that the residual fat content from Run 1 was as high as .5 to .6%, the contact time was changed from 40 to 60 minutes. After it was observed that this change did not make any appreciable difference in the residual fat content, the conditions for Run 3 were shifted back to those of Run 1. The solvent ratio of 1.9:1 was considered exceptionally high for this extractor, and the solvent entered just below its boiling point.

Considerable difficulty was encountered in recovering the solvent from the extracted yeast. A three-pass continuous tunnel dryer was used for this purpose. When it was observed that the yeast was getting a toasted flavor from the drying operation, the

steam pressure on each of the passes was reduced from 20 to 15 p.s.i.g.; the reducing of the pressure below this point resulted in traces of the solvent remaining with the extracted yeast, which obviously could not be tolerated for a food product. Because the process was continuous, the contact time in the dryer was fixed by the extraction conditions and was about six minutes. This difficulty would indicate that the extracted yeast must be dried under very careful drying conditions, either with much less heat at a slower speed or in a continuous vacuum dryer, preferably the latter.

Analyses of the Extracted Fats. The literature is abundant in information on the composition of the fatty acids of yeast fat (1, 4, 5, 6, 7, 8, 9). As is to be expected, even on the same strain of yeast, *Saccharomyces cerevisiae,* there is considerable difference reported in the composition of the fatty acids, especially the minor constituents. This is largely due to differences in environment during fermentation. The literature seems to be in agreement that the primary saturated fatty acids of yeast fat are palmitic and stearic, with palmitic being predominant, and the primary unsaturated fatty acids are oleic and linoleic, with oleic acid being considerably more predominant than linoleic. Rewald (10) has reported that the phospholipid concentration in yeast fat extracted by a mixture of benzene and alcohol from undebittered brewers' yeast is about 45.5% whereas Newman and Anderson (11) report that about 17.5% of the fat extracted from fresh moist *Saccharomyces cerevisiae* is phospholipids and that the composition of these phospholipids is approximately four parts lecithin to one part cephalin. The glycerol content of the fat has been reported by Täufel, *et al.* (6) as 5.3%, and the same authors report that the unsaponifiable fraction is 19.6%, consisting of 3.3% ergosterol and cryptosterol and 16.3% squalene. Holmberg (1) however reports that the unsaponifiable fraction is only 3.1%.

The analyses of the yeast fats reported in Table IV were run on a dried, non-viable, primary grown, food yeast, specifically *Saccharomyces cerevisiae,* using official A.O.C.S. methods. For comparative purposes the fat content (petroleum ether extractables) of the yeast before and after extraction is given. It should be pointed out that the analyses were not performed until about 18 months after extraction and that this may account for the high free fatty acids content.

TABLE IV

Analysis of Fat Extracted from Yeast

	Horizontal Extractor	Vertical Extractor
Moisture	.74%	3.27%
Free Fatty Acids	6.4	20.0
Solid Fatty Acids	17.38	18.95
Liquid Fatty Acids	82.62	81.05
Saponification Number	142.0	141.5
Iodine Number	115.3	135.2
Unsaponifiable Matter	8.96	9.85
Acetone Insoluble Matter, "Phospholipids"	47.10	18.39
Benzene Insoluble Matter	.34	.32
Fat in Unextracted Yeast (as is)	1.7	1.0
Fat in Extracted Yeast (as is)	.033	.45

The fat is very dark brown in color and viscous and has a strong yeast-like odor and taste. Upon standing it shows no tendency toward rancidity. Conventional methods for refining were unsuccessful because of the high free fatty acids content and high unsaponifiable and high phosphatide fractions.

Neuss (2) reported in 1916 that the fat makes a good soap curd but the fat which was obtained in our work could not be considered a good soap stock by today's standards because of its dark color and high unsaponifiable fraction.

In certain foods, particularly bread, it is sometimes desirable to have yeasty flavor, and since the fat had concentrated the yeast odor and flavor, it was decided to try the addition of yeast fat to bread. Baking tests were conducted in an experimental bakery using .2% crude fat, which was added at the remix, and this was compared with control samples not containing the fat. It was observed for the sample with the fat that the dough characteristics were normal at the bench and moulder and that proof time was slightly shorter and total dough CO_2 slightly greater. The loaf volume however was 100 cc. less for the bread with fat added, and the fat seemed to have a binding effect on the dough; there was no appreciable difference observed in aroma or flavor.

The fat may have some value for its ergosterol content (vitamin D percursor), but this possibility was not investigated. In addition, the high lecithin content may be of some interest commercially.

Organoleptic Tests on Extracted Yeasts. Samples of the extracted yeast from the horizontal extractor runs were submitted to a trained taste panel immediately after extraction and compared with unextracted yeast. For this purpose one gram of yeast was dispersed in 100 cc. of tomato juice. The preference was overwhelming in favor of the extracted yeast. Another advantage of the extracted yeast was its lighter color.

This same yeast was submitted to a different, trained taste panel four and five months after extraction and compared with fresh dried unextracted yeast and No. 105 yeast. The latter was an especially bland yeast produced by a special laboratory fermentation process.

The taste tests were conducted by the following procedure:

Each dried yeast was blended in the ratio of 6 g. to 56 g. of Nestle's Instant Cocoa Drink. The total amount of mix (62 g.) was suspended in 960 g. of homogenized milk, using a Waring Blendor for five minutes to obtain better dispersion.

The cocoa beverages were served at room temperature in one-ounce glasses, marked in code. Printed directions were given to the tasters, who rated the beverages in descending order of preference. Plain white bread, with crust removed, was used between samples to "clear the taste." The results of these tests are given in Table V.

TABLE V

Duplicate Preference Taste Tests

First Test (Four Months after Extraction) (40 Persons Participating)

	Unextracted Yeast	Extracted Yeast	No. 105 Yeast
First Preference	17.5%	15.0%	67.5%
Second Preference	55.0%	22.5%	22.5%
Third Preference	27.5%	62.5%	10.0%

Second Test (Five Months after Extraction) (40 Persons Participating)

	Unextracted Yeast	Extracted Yeast	No. 105 Yeast
First Preference	32.5%	15.0%	52.5%
Second Preference	40.0%	25.0%	35.0%
Third Preference	27.5%	60.0%	12.5%

The personnel of the taste-testing group fluctuates somewhat, due to absences, illness, conflicts in schedules, etc. Thirty-one of the same individuals participated in the yeast preference test on both dates. Of this number, only 10 people consistently reproduced their judgments.

These results indicate that, although the taste of the extracted yeast immediately after extraction is preferable to the unextracted yeast, upon storage some change occurs in the extracted yeast which makes it undesirable from a taste standpoint.

Taste tests on the yeast from the vertical extractor runs indicated that even immediately after extraction the taste of the extracted yeast was unsatisfactory. This can be explained by: a) only about one-half of the fat was removed from the yeast, and b) the yeast was toasted in the drying process.

Conclusions

The pilot studies performed on the extraction of fat from yeast indicate that it is possible commercially to extract yeast to .05% residual fat or less, using finely pulverized yeast in a continuous countercurrent horizontal extractor and Skellysolve B for the solvent.

The yeast obtained by this extraction is lighter in color and, when fresh, has a blander taste than similar unextracted yeast, provided that it is dried under carefully controlled conditions so that all the solvent is removed and yet the yeast is not toasted. However after several months' aging, the extracted yeast does not compare favorably with fresh unextracted yeast on a taste basis.

The fat extracted has a a high phosphatide and high unsaponifiable fraction and will vary considerably in composition, depending on species of yeast, environment during growth, and method of extraction. Because of its composition it cannot be conveniently refined and therefore would not compete favorably with vegetable oils for bulk industrial use.

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Advances in Protective Coating Chemistry: Fatty Acids, Polymerization, End Products[1]

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ONE cannot discuss advances in protective coating chemistry without pointing out that the technology involved is very old. In ancient times men were already aware of the desirability of protecting wood or metal surfaces with a film of some sort of material that would resist scratching and abrasion. Some of these ancient coatings are still extant today, and the scientific investigation of these allows the imagination to call forth a picture of an ancient Egyptian who has come upon some soft, naturally occurring resins. Today these might be called amber, sandarac, or mastic. He also had available to him certain oils which probably were not too remote from linseed oil. The Egyptian heated the resin and the oil together so that there resulted a sort of varnish which he could apply, while it was still warm, with his finger or with a knife. Today in various museums there are mummy cases coated with the varnish prepared by that ancient Egyptian four to five thousand years ago. And the Egyptian paint technologist would have ample reason to be very proud of his work for the varnish is frequently still bright and its surface undamaged and unmarred.

Considerably more is known about the composition of the protective coatings of the Middle Ages. Although many of these very early formulations were shrouded in secrecy, some have been preserved. And it is perhaps with some amusement that we note one technologist of some centuries previous who recommended that a certain type of varnish be cooked by allowing the oil to burn at the mouth of the kettle for a period of time sufficient for the recitation of three Pater Nosters.

These facets of protective coating history are offered here only to show that the protection and decoration of surfaces has been the concern of man since the dawn of civilization. In this paper will be discussed protective coatings which dry by polymerization; that is, coatings which, when spread out on a surface, polymerize—under the influence of air, light, heat, or chemicals—to yield solid, resistant, continuous films. Such coatings are based largely on compounds formed from unsaturated fatty acids which, as we shall see, provide the functionality necessary for polymerization and subsequent film formation. The second large class of coatings, which are not to be discussed here, are those based on cellulose and other polymers. These are already sufficiently highly polymerized so that they provide films as soon as they are applied to a surface.

As already indicated, fatty acids are the basic building blocks of polymerizable protective coatings.

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