Growth Studies of Yeast in Refined Syrups at Various Levels of Solids and Nutrients

M. Bolinder *

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INTRODUCTION

Yeasts are microorganisms which have the ability to utilize most common sugars in their metabolic process. The alcohol in wine is a familiar by-product of yeast metabolism using sugar. Under certain conditions yeast may grow in stored sugar syrups. Yeast growth when making wine is desirable but undesirable when growth occurs in stored sugar syrups.

As a supplier of blended syrups, it is desirable to know in which of certain blends yeasts may be a problem during storage.

The resistance of sugar syrups to yeast spoilage is highly dependent on two factors; first, the amount of water present and second, the availability of nutrients. This article deals with tests made at Spreckels where yeast growth was monitored in syrups at various levels of solids and nutrients.

MATERIALS AND METHODS

Tests were made by preparing sugar syrups with varying solids levels with and without added nutrients. One test was made where the solids level remained constant while the nutrient level varied.

Various prepared syrups were inoculated with two osmotolerant yeasts, *Saccharomycess rouxii* ATCC22027, and an unidentified yeast isolated from fermenting 28 DE corn syrup. The nutrients added were Difco brand Yeast Nitrogen Base. Yeast Nitrogen Base (YNB) contains all the essential nutrients and vitamins necessary for the cultivation of yeast except a carbohydrate source(1).

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* Central Testing Laboratory Supervisor, Amstar Corporation, Spreckels Sugar Division, Woodland, CA 95695.

Ten milliliters of each prepared syrup was placed in separate 16x150 mm culture tubes. Approximately three milliliters of paraffin wax was placed on the syrup surface forming a plug in each culture tube. The culture tubes were incubated at 30°C. Yeast growth was monitored by periodic observation of the wax plug moving upwards as gas formed by fermentation.

RESULTS

Test 1

The first test consisted of observing yeast growth in some blended syrups ranging from 71.0 to 78.3% solids.

The blends were prepared using varying amounts of sucrose, 50% inverted syrup, 62 DE corn syrup, and HFCS.

Table 1. Saccharide composition and solids level of blends used for yeast growth studies.

Blend No.	Parts per hundred, Solids Basis Sucrose	50% Invert	62 DE Corn Syrup	HFCS	% Solids
1	50		25	25	71.0
2	60		40		72.0
3	55		45		72.8
4	50		45	5	72.8
5	5		65	30	77.7
6		70	25	5	77.9
7	1		65	34	77.9
8		1	65	34	78.0
9		50	40	10	78.3

To optimize the yeasts growing conditions, 670 ppm YNB was added to one series of blends.

Table 2. Milliliters of CO_2 produced at various incubation intervals in nine blends both with and without added nutrients.

Blend	% Solids	Water Activity	Days Incubation 1	3	6	7	8	9	10	13	14	15	16	17	22	27	34
1	71.0	.86	0	0	0	0	0	0	1	2	2	2	2	3	3	4	5
1+YNB	71.0	.86	0	0	1	1	1	L	L	L	L	L	L	L	L	L	L
2	72.0	.87	0	0	0	0	0	0	1	1	2	2	2	2	3	5	6
2+YNB	72.0	.87	0	0	0	1	1	2	2	L	L	L	L	L	L	L	L
3	72.8	.87	0	0	0	0	0	0	0	1	1	2	2	2	2	2	2
3+YNB	72.8	.87	0	0	0	0	0	1	2	3	3	3	3	3	3	3	3
4	72.8	.86	0	0	0	0	0	0	0	1	1	2	2	2	2	3	3
4+YNB	72.8	.86	0	0	0	0	0	1	1	1	2	2	2	2	2	2	2

Table 2. continued.

Blend	% Solids	Water Activity	Days Incubation 1	3	6	7	8	9	10	13	14	15	16	17	22	27	34
5	77.7	.80	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
5+YNB	77.7	.80	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
6	77.9	.79	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
6+YNB	77.9	.79	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
7	77.9	.80	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
7+YNB	77.9	.80	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
8	78.0	.79	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
8+YNB	78.0	.79	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
9	78.3	.79	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
9+YNB	78.3	.79	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0

L = Gas leaked around side of wax plug.

The results of the first test indicated yeast growth is closely related to the level of solids and nutrients. No yeast growth occurred in any blend at solids levels exceeding 77.0%. At lower solids levels that yeast growth was more rapid in those tubes with added nutrients.

A value related to solids level and carbohydrate makeup which should more closely reflect the propensity of a syrup to yeast spoilage is water activity (A_W). Water activity defines the effective concentration of water as a reactant (2). Water activity may be approximated using the following formula(3):

$$A_W = \frac{\text{moles water}}{\text{moles water + moles solute}}$$

The water activity of each blend tested was calculated. These values are listed in Table 1. A hiatus in the A_W of blends tested appeared between .80 and .86.

Test 2

Another test was set up to check yeast growth at A_W .80 to .86. Dilutions of 50% inverted syrup were made in the range of 68 to 77% solids and enriched with 1000 ppm yeast nitrogen base. These were set into culture tubes as before.

The tests indicated yeast growth was very time retarded approaching 76% solids (.81 A_W). At 77% solids (.80 A_W) no growth was evident for 34 days.

Table 3. Incubation time in days until at least 1 milliliter gas producted in culture tubes with 50% inverted syrup at concentrations from 68-77% solids.

A_W	% Solids	Days Required to develop 1 mil gas
.86	68	4
.84	72	8
.82	74	14
.81	76	28
.80	77	>34

Results from Test 1 indicated yeast activity was greater at elevated nutrient concentrations. This suggested that at low nutrient concentrations yeast may not be able to proliferate. A third test was set up to determine the effect of nutrients on yeast growth.

Test 3

This test used a highly refined HFCS diluted to a solids level of 50% (A_W = .91) with yeast nitrogen base added at levels varying from 0 to 500 ppm.

Table 4. Incubation time until at least 0.5 milliliter gas produced in culture tubes with 50% solids HFCS at varying nutrient levels.

PPM Added YNB	Time Required to develop 0.5 ml gas
0	> 5 months
5	> 5 months
10	> 5 months
25	93 days
50	14 days
100	2 days
500	2 days

The data indicate no yeast growth occurred at 10 ppm added yeast nitrogen base or less. Yeast growth was appreciably retarded at 25 ppm added YNB. Growth occurred rapidly at 100 and 500 ppm added YNB. High fructose corn syrup normally contains about 40 ppm nitrogenous material (4). Yeast growth would be very retarded at a level approaching 60 ppm nitrogenous nutrients.

DISCUSSION

For 50% inverted syrup with nutrients added, a solids level of about 77% is necessary for safe prolonged storage. This is a water activity of 0.80. Troller and Christian found the preservation of jams and jellies relies on attaining A_W levels of about 0.80 or lower(2). In actual practice the nutrient level of some highly refined carbohydrates would be quite low thus allowing safe storage at somewhat lower solids levels than indicated by the experimental nutrient enriched syrups. Those nutrients normally found in sugar syrups would probably not be as "balanced" as those used for the tests.

Blends containing unmixed corn syrup and sucrose syrup are especially prone to spoilage. This is because the sucrose syrup contributes water and the corn syrup contributes nitrogenous nutrients to the blend. Many unmixed corn syrups normally contain in excess of 1000 ppm nitrogenous substances (4).

Samples of 80% solids 42 DE corn syrup have been observed actively fermenting in the laboratory. One could expect yeast growth since the A_W of 42 DE syrup at 43 Baumé is 0.85 and ample nutrients for yeast growth are present.

The solids level necessary to obtain a A_W of 0.80 for some common syrups follow:

Table 5. Calculated % solids at 0.80 A_W for some common sweeteners.

	% Solids
Sucrose syrup	82.
50% inverted syrup	77.
HFCS 42	73.
28 DE Corn Syrup	90.
42 DE Corn Syrup	86.
62 DE Corn Syrup	80.

For sucrose syrup to have a A_W of .80, a solids level of 82% is required. This is prohibitively high since sucrose would crystallize at normal temperatures. Fifty percent inverted syrup is safe from spoilage at the normal solids level of 77.0%. High fructose corn syrup is usually safe because it is highly refined and is normally sold at a solids level near .80 A_W. Low conver-

sion corn syrups are quite likely to spoil if given enough time. At 62 DE, corn syrups should be safe from spoilage at normal solids levels exceeding 81.6% (43 Baumé).

SUMMARY

A syrup may be considered safe from yeast spoilage if the water activity is less that 0.80 or the syrup contains less than 60 ppm total nitrogenous nutrients.

LITERATURE CITED

(1) Formula information from bottle label of Yeast Nitrogen Base, Difco Laboratories, Detroit, Michigan.

(2) Water Activity and Food; 1978, Troller, John A., & J. H. B. Christian, Academic Press.

(3) Bone, D,; 1973, Water Activity in Intermediate Moisture Foods, Food Technology, 27:71.

(4) Unpublished laboratory data from Spreckels Corn Operations.