Research Note

EVALUATION OF A COMMERCIAL YEAST NUTRIENT FOR HIGH TEST MOLASSES FERMENTATION[1]

The availability of locally produced blackstrap molasses (BM), the main raw material for rum production, has been declining during the last decade. Consequently, most BM has to be imported. A vigorous local rum industry should not depend on importing its principal raw material. Production of high test molasses (HTM) has been proposed as an alternative to ensure raw material for the Puerto Rican rum industry.

HTM is defined as a clarified sugar cane syrup, partially inverted to prevent crystallization, and evaporated to 85° Brix. A HTM manufacturing procedure was developed at the Rum Pilot Plant.[2] A process for alcoholic fermentation with HTM was developed.[3,4] Distillates with more esters and less fusel oil, convenient for high quality rum manufacture, were obtained.[5]

It is known that fermentation rate is proportional to the amount of live yeast cells present. During the course of fermentation, part of the initial inoculum is replaced by new yeast cells. The substrate should provide the nutritional requirements that yeast as living entities need for this replacement. It has been experimentally determined that HTM as fermentation substrate requires additional nutrients to achieve these nutritional conditions. Yeast extract, a dehydrated form of the water soluble portion of autolyzed yeast, rich in naturally occurring B-complex vitamins, enhanced fermentation of HTM.[3] Its high cost limits its widespread use in the fermentation industry. A more economical alternative was sought.

Yeastex 61[6] is a commercial preparation by Scott Laboratories, Inc. which has been approved for use in wineries by the US Bureau of Alcohol Tobacco & Fire Arms. It is a mixture of mineral salts and organic nutrients essential for vigorous yeast growth with important trace elements, amino acids and other growth factors. Similar preparations are available from other companies. Its use in HTM alcoholic fermentations was investigated as an appropriate economical nutritional environment in HTM mashes. Besides Yeastex 61, sources of nitrogen [$(NH_4)_2SO_4$] and phosphorus ($NH_4H_2PO_4$) were studied.

The general fermentation procedure consisted in diluting the HTM to a wort of

[1]Manuscript submitted to Editorial Board 7 April 1987.

[2]Rosado, E., 1983. Método para la elaboración de melaza rica. Progress Report 23-83, Rum Pilot Plant, Agric. Exp. Stn. Univ. P. R., Río Piedras, 1-13.

[3]Murphy, N. F., 1984. Fermentation of high test molasses. *J. Agric. Univ. P. R.*, 68 (1): 33-44.

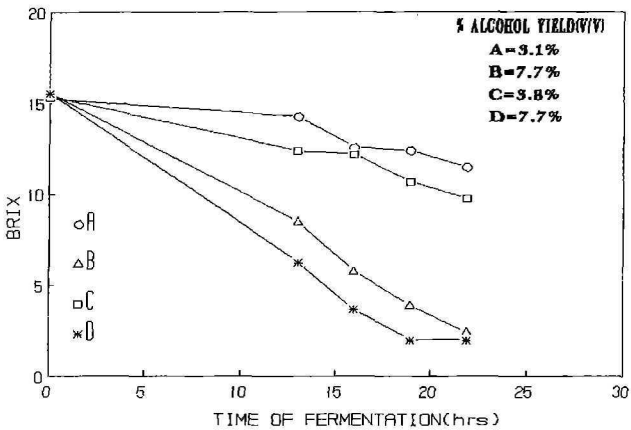
[4]Cacho, E., N. F. Murphy and E. Fontanet, 1986. Pilot Plant fermentation of high test molasses compared with blackstrap molasses using factorial design. *J. Agric. Univ. P. R.* 72 (1): 9-17.

[5]Martínez, G. and N. Murphy, 1984. Congeners from high test molasses alcoholic fermentation. *J. Agric. Univ. P. R.* 68 (1): 59-65.

[6]Trade names are used solely for the purpose of providing specific information. Mention of trade names does not constitute a guarantee or warranty by the Agricultural Experiment Station of the University of Puerto Rico or an endorsement over other preparations not mentioned.

optimum sugar concentration (17.2° Brix), adding the desired nutrients, pasteurizing at 170° F for 10 min and inoculating with a vigorous growing yeast seed of *Saccharomyces* strain PPR 80. The seed was prepared by succesive transfers to larger volumes as described in fermentation of high test molasses.[1] Initial mashes were analyzed for total acidity and total sugars by described methods.[2] Fermentations were conducted in triplicate in 20-liter glass fermenters. Total mash volume was 16 liters, two liters of which were yeast seed. Periodic °Brix readings of the mash were recorded as a measure of rate of fermentation for 22 hours. The final fermented mashes were analyzed for °Brix, pH, total acidity, residual sugars and alcohol.

An initial study with the manufacturers' recommended concentration for wine (0.25 g/L) was performed and compared with $(NH_4)_2SO_4$-enriched mashes. Figure 1 shows that both Yeastex 61 and $(NH_4)_2SO_4$ improved yeast performance when used individually, but better results were obtained when both nutrients were used concurrently. Experiments were then conducted with higher concentrations of Yeastex 61 since faster rates of fermentations are desirable for the rum industry than for wine manufacture. Table 1 and figure 2 present the results. Note that residual sugars were lower and alcohol yields were higher in fermentations containing Yeastex 61. Evidently, the preparation added nutrients required by the yeast for optimum fermenta-



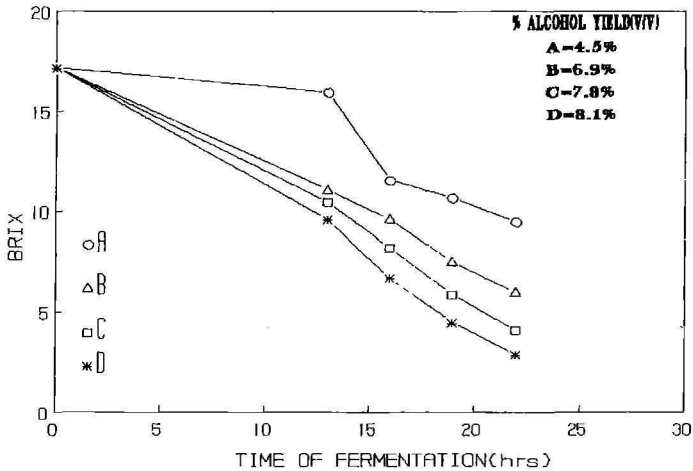
A: no nutrients added; B: $(NH_4)_2SO_4$ = 1.5 g/L; C: yeastex 61 = 0.25 g/L; D: yeastex 61 = 0.25 g/L, $(NH_4)_2SO_4$ = 1.5 g/L

FIG. 1.—High test molasses fermentation, effects of additional nutrients.

[1]Official Analytical Methods of the Rum Pilot Plant, 1969. Agric. Exp. Stn., Mayagüez Campus, Univ. P. R., Río Piedras, P. R.

TABLE 1.—*Effect of nutrients on fermentation of high test molasses by yeast strain 80 PPR*

	Nutrients added																
Yeastex g/liter	0	0	0	0	0	1.0	1.0	1.0	1.0	2	2	2	2	3	3	3	3
$NH_4H_2PO_4$ g/liter	0	0	0.5	1.0	1.5	0	0.5	1.0	1.5	0	0.5	1.0	1.5	0	0.5	1.0	1.5
$(NH_4)_2SO_4$ g/liter	0	1.5	1.5	1.5	1.5	1.5	1.5	1.5	1.5	1.5	1.5	1.5	1.5	1.5	1.5	1.5	1.5
	Brix readings																
Hour																	
0	16.1	15.8	15.7	15.7	15.9	16.0	15.8	16.0	15.9	15.8	15.8	15.7	15.7	15.9	15.7	16.0	15.8
13	132.0	11.1	10.5	10.9	11.3	9.6	9.8	10.2	10.2	10.1	9.7	8.7	8.9	9.5	9.3	9.5	9.8
16	11.6	9.6	8.2	8.7	9.0	6.7	6.7	7.2	7.1	7.0	6.3	5.2	5.4	6.2	6.1	6.4	7.0
19	10.7	7.5	5.9	6.3	6.7	4.5	4.2	4.3	4.1	4.4	3.8	3.0	3.1	4.0	3.9	4.0	4.5
22	9.5	6.0	4.1	4.2	4.9	2.9	2.8	2.8	2.9	3.0	2.8	2.7	2.7	2.9	2.8	2.8	2.8
	Samples at 22 hours fermentation																
Sugar																	
g/100 ml	8.5	3.6	1.5	3.4	2.1	0.8	0.7	0.7	0.7	0.8	0.7	0.7	0.7	0.7	0.7	0.7	0.7
% alcohol	4.5	6.9	7.8	7.7	7.4	8.1	8.1	8.2	8.1	8.2	8.1	8.1	8.0	8.0	8.0	8.0	8.0



A: yeastex = 0, $NH_4H_2PO_4$ = 0, $(NH_4)_2SO_4$ = 0; B: yeastex = 0, $NH_4H_2PO_4$ = 0, $(NH_4)_2SO_4$ = 1.5 g/L; C: yeastex = 0, $NH_4H_2PO_4$ = 0.5 g/L, $(NH_4)_2SO_4$ = 1.5 g/L; D: yeastex = 1.0 g/L, $NH_4H_2PO_4$ = 0, $(NH_4)_2SO_4$ = 1.5 g/L

FIG. 2.—High test molasses fermentation, effects of additional nutrients.

tion. Concentrations of 2.0 g/L and 3.0 g/L exhibited faster rates up to 19 hours fermentation time; but by completion time, 22 hours, 1.0 g/L Yeastex 61 produced equivalent results in terms of sugar consumed and alcohol yield. The use of ammonium phosphate (phosphorus source) appears to be unnecessary as Yeastex 61 provided the adequate amounts of phosphorus needed. On the basis of cost-benefit analysis, we recommend a combination of Yeastex 61 (1 g/L) with ammonium sulphate (1.5 g/L) as the best alternative.

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