SEARCH CORPORATION OF THAILAND

in collaboration with
DEPARTMENT OF SCIENCES, MINISTRY OF INDUSTRY

COOPERATIVE RESEARCH PROGRAMME NO. 57
UTILIZATION OF GLUTINOUS RICE

RESEARCH PROJECT NO. 57/15
ALCOHOLIC FERMENTATION OF GLUTINOUS RICE

REPORT NO. 1
A PRELIMINARY STUDY OF ALCOHOL YIELDS FROM GLUTINOUS RICE AND OTHER CARBOHYDRATE SOURCES

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ASRCT, BANGKOK 1972
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F O R E W O R D

Thailand at times has big surpluses of rice especially the glutinous rice. The Ministry of Economic Affairs estimated that there would be a total surplus of 700,000-800,000 tonnes in B.E. 2514. The Government, in order to help solve this problem, has asked the Department of Science, Ministry of Industry and ASRCT to carry out a joint effort to increase the utilization of glutinous rice. By joint agreement between the two organizations, ASRCT undertook a study on alcoholic fermentation of glutinous rice. Since there are other starchy materials produced in large quantities in the country, they were also included for comparison. From the result of the preliminary investigation there appear to be several kinds of raw materials which could be used with advantage in the production of alcohol by the distilleries. It is hoped that the effort would arouse the interest of the manufacturing sector to widen the choice of the raw materials employed and to improve the conventional method of production for the benefit of the consumers and perhaps for a better opportunity of exporting the finished products.

A PRELIMINARY STUDY OF ALCOHOL YIELDS FROM GLUTINOUS RICE AND OTHER CARBOHYDRATE SOURCES

By Sman Vardhanabhuti*, Jiraporn Sukhumavasi*, and Pravat Lauhasiri+

SUMMARY

Preliminary study on the yield of alcohol was made with ordinary white rice, white glutinous rice, black glutinous rice, raw cassava, sorghum, corn, and molasses. The starchy materials were converted into fermentable sugars by commercial enzymes prior to the yeast fermentation. For the production of industrial alcohol by local distilleries under their existing conditions, molasses were found to be still the substrate of choice, but dry cassava would be another choice when molasses was in short supply. For beverage alcohol, sorghum and broken rice (both ordinary and glutinous) could be used with advantage. Corn and undermilled rice (both kinds) were found to be far too expensive for industrial alcohol production. Alcohol made from black glutinous rice has a special aroma peculiar to itself and may be useful for the production of a unique type of rice wine (spirit). Starch conversion by commercial enzymes is not practiced by the local industry and needs further study.

INTRODUCTION

Molasses is the main substrate used in alcoholic fermentation in this country. Broken glutinous rice is used only for the production of glutinous rice spirit specially favoured by the Chinese community, and the production is small. Sorghum is used for the production of kaoliang wine, formerly imported from Hong Kong and Taiwan. There is only one Chinese company producing the wine occasionally whenever the market demands it. Molasses does not need conversion and is fermented directly to alcohol by the yeast. Glutinous rice and sorghum are converted into fermentable sugars by mold brans produced by the distillery itself by

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traditional method. The percentage of conversion by traditional mold bran is quite low. Commercial enzymes have never been used by the industry. In the present study commercial enzymes were used and a wide range of raw starchy substrates as well as molasses were tested. Well standardized methods of mash production, alcoholic fermentation, and estimation of alcohol yield have been adopted. The following are the results of the investigations.

MATERIALS AND METHODS

a) Materials

i) Substrates

Nine substrates were tested. These were cassava root, corn, whole sorghum, undermilled ordinary rice, undermilled black glutinous rice, broken ordinary rice, broken white glutinous rice, and molasses.

Undermilled white glutinous rice was milled from paddy at the Department of Agriculture, Ministry of Agriculture.

The undermilled black glutinous rice was bought from a mill in Thanyaburi.

The Mahakhun Distillery Co., Ltd. at Bang Yi Khan, Thon Buri kindly supplied the broken white glutinous rice. Molasses was kindly provided by the Ayutthaya Alcohol Distilleries.

The other substrates were bought from local markets.

ii) Enzyme preparations

Two commercial enzymes, Maxamyl for starch liquefaction and Maxydrase for starch conversion, were obtained from the Royal Netherlands Fermentation Industries Limited, Delft, Holland, as commercial samples without charge.

iii) Yeast

Saccharomyces carlsbergensis was kindly provided by the Ayutthaya Alcohol Distilleries.

b) Methods

i) Preparation of raw materials

Fresh cassava roots were washed and cut into chips without peeling. The chips were sun-dried and the loss of weight through drying was recorded.

Each of the raw substrates was ground by a disc mill (Karl Kolb, type KG 48, Frankfurt/M, Germany) and sieved through a U.S. standard sieve No. 50 (48 mesh). An aliquot of each was sent for chemical analysis.

ii) Starch liquefaction and conversion

The method of starch liquefaction and conversion was modified from Technical Data Sheet No. 652 of the Chemical and Biochemical Division, the Royal Netherlands Fermentation Industries Limited. The tests were performed in duplicate.

Flour suspension was made, using 253.3 g of flour in 927 ml of tap water. The pH of the suspension was adjusted to 7.0 with sodium hydroxide solution. (Adjustment of pH using calcium carbonate as recommended by the producer was not satisfactory in our case because it took very large amounts of calcium carbonate to raise the pH to the desired level. Moreover in the case of sorghum, which took 610 g, the large amount of calcium carbonate greatly depressed the activity of the yeast in subsequent alcoholic fermentation.)

Maxamyl (0.0406 g for cassava and broken rice, 0.0812 g for the others) was made pappy with water and added to the flour suspension in a glass beaker. The level of the fluid was marked. With constant stirring, the mixture was heated in a water bath until it completely gelled. The temperature of the water bath was then kept constant at 75°C. Further stirring at this temperature resulted in progressive liquefaction. After complete liquefaction the temperature of the mash was raised quickly to 90°C and kept at that level for 20 minutes to inactivate the enzyme. The mash was then cooled, and tap water was added up to the original marked level. The pH of the liquefied starch was adjusted to 4-4.5 with concentrated hydrochloric acid. Maxydrase (0.5066 g) was then added, the mixture well stirred, and the conversion was allowed to take place at 55°C in a water bath for four days with occasional stirring. Fresh tap water was added to the cook at times to compensate for the amount lost through evaporation. After conversion a 56-ml aliquot (the extra 5% amount originally included in the mash for sampling purpose) of each converted cook was taken out for chemical analysis.

iii) Preparation of yeast inoculum

One loopfull of an 18-hour old potato dextrose agar slant culture of Saccharomyces carlsbergensis was inoculated into 10 ml of sterile 16° Balling malt extract (Difco) in a test tube and kept at 35°C in an incubator. Two successive transfers in the same medium were made once every 24 hours. The whole 10 ml of the last subculture was transferred into 200 ml of the same malt extract medium in an Erlenmeyer flask and the flask was kept at 35°C for 20 hours to be used as an inoculum for alcoholic fermentation of the cook (the converted substrate).

iv) Fermentation

The fermentation procedure was modified from the process described by Stark et al. (1943). It was performed under sterile conditions.

Cooled converted cook was poured into 2-litre graduate cylinder. Sterilized stillage (300 ml), obtained from the Ayutthaya Alcohol Distilleries, were screened through cheese cloth and added to the cook, which was then mixed thoroughly. The pH of the cook was adjusted to 4.8 with 1 N sulphuric acid if necessary. The volume was made up to 1500 ml with sterile tap water. The content of the cylinder was mixed by pouring back and forth from the cylinder to a beaker twice. The pH was again checked; if it was above 4.8, it was set back to 4.8 with a few drops of 4 N sulphuric acid.

Aliquot (7 ml) of the yeast inoculum (2% of the total mash volume) was added to each of 500-ml JENA-glass, widemouthed, rubber-stoppered reagent bottles, which served as the fermenters. A 500-ml graduate cylinder was rinsed with 50 ml of the mash, and 350 ml of the latter was measured out and poured into the fermenter, allowing it to drain until there was only 1 drop every 2 seconds. Three fermenters were used for each cook, and a separate graduate cylinder was used in every case. The trap was then attached to the fermenter. It was prepared as follows:

Glass beads and 2 ml of mercury were placed to a depth of about 1.25 cm in a 25 mm x 200 mm test tube. Distilled water (50 ml) was then added. A bent glass tube extending to the bottom of this trap was connected with the fermenter through a rubber stopper.

The fermenters were incubated at room temperature (25-30°C) for the first 20 hours, at 35°C in a constant temperature water bath for the next 30 hours, and at 37°C for the final 16 hours. This is a modification of the original method to fit with the local ambient temperature. At least twice during the fermentation, i.e. at 20 and 50 hours, the fermenters were shaken thoroughly by swirling until all the mash adhering to the bottom of the flask was insuspension.

For molasses, a proper amount of known reducing sugar content was diluted to 1500 ml to make a solution of 12% reducing sugar. The fermentation was then carried out in the usual manner.

The leftover cook was centrifuged at 2000 rev/min for 10 minutes in a Junior 111 Chriss Centrifuge (Karl Kolb). The supernate was collected and sent for chemical analysis.

v) Distillation

At the end of the fermentation, the mash was mixed thoroughly, the fermenter and its content weighed, and the weight of the mash calculated. Exactly one half of the mash was weighed into a one-litre distilling flask and 50% of the water in the trap was then added to it. The mash was distilled into a 100-ml volumetric flask containing 5 ml of distilled water prior to distilling. The collecting end of the condenser was connected with a glass tip so that all the vapour passed through the water in the receiver. Iced water was used to cool the condenser, and the receiver was immersed in an ice bath.

After the distillate was up to the neck of the collecting flask, the distillation was stopped. The flask was warmed to 25°C, and the distillate was made up to volume (100 ml) with distilled water. The percentage of the alcohol was then measured by an immersion alcoholmeter (Richter and Tralles, Karl Kolb). The remainder of the fermented mash was centrifuged at 2000 rev/min for 10 minutes, and the supernate was collected and sent for chemical analysis.

vi) <u>Chemical analysis</u>

All chemical analyses were performed by the Analytical Unit, Chemical Technology Group, TRI. The methods used were those given in the AOAC manual (Horwitz 1965).

The raw substrates were analysed for the contents of moisture, starch, ash, reducing sugar, total nitrogen, and fat.

The converted cooks were analysed for the contents of fat, starch left, total sugar, reducing and non-reducing sugar.

The mashes before and after fermentation were tested for pH titrable acidity, and per cent reducing sugar.

The yeast mash and the stillage were analysed for reducing sugar content.

vii) <u>Calculations</u>

% Conversion based on starch

$$= \frac{\text{g of starch converted in 100 ml mash}}{\text{g of initial starch in 100 ml mash}} \times 100$$

% Conversion based on glucose

$$= \frac{\text{Actual g of glucose obtained in 100 ml mash}}{\text{Theoretical g of glucose from starch in 100 ml mash}} \times 100$$

% Fermentation efficiency based on starch

$$= \frac{\text{Actual g of abs. alcohol obtained from 100 g raw substrate}}{\text{Theoretical g of abs. alcohol from 100 g raw substrate}} \times 100$$

Theoretical yield of alcohol = % starch in flour x factor for changing starch to glucose (10/9) x factor changing glucose to abs. alcohol (0.511)

% Fermentation efficiency based on sugar fermented

$$= \frac{\text{Actual g of abs. alcohol obtained from 100 g raw substrate}}{\text{Theoretical g of abs. alcohol from glucose in 100 g raw substrate}} \times 100$$

Proof gallon per bushel (wet basis)

$$= [\% \text{ v/v of alcohol in distillate} - (\text{initial sugar concentration of yeast mash, \% w/v, as maltose}) \times 0.0208] \times \frac{100 (\text{ml of distillate})}{175 (\text{ml of mash distillate})} \times \text{concentration of mash } (38 \text{ gal/bu}) \times \frac{2}{100}$$

$$= \% \text{ v/v of alcohol in distillate} - (\% \text{ w/v initial sugar of yeast mash} \times 0.0208) \times 0.4343$$

Proof gallon per bushel (dry basis)

$$\frac{\text{Proof gallon per bushel (wet basis)}}{\text{Per cent of dry grain mashed}} \times 100$$

(100 - per cent moisture of grain mashed)

RESULTS

The chemical composition of various raw materials is given in Table 1. Their starch contents range from the highest to the lowest in the following order: broken ordinary white rice, broken white glutinous rice, undermilled white glutinous rice, undermilled ordinary rice, cassava and undermilled black glutinous rice, corn and sorghum. It is of interest to note that undermilled black glutinous rice has a very high total nitrogen content (2.1%), distinctly higher than that of corn and sorghum.

The amounts of enzymes used for liquefaction and conversion of the raw starches are given in Table 2.

Table 3 and Table 4 give respectively the results of enzymatic conversion of various substrates on the one hand and the composition of the mashes before and after fermentation on the other. Very good conversion was obtained with cassava and the rices, good with sorghum, and slightly less so with corn.

The yields of alcohol are given in Table 5. The highest yield of alcohol was obtained from raw cassava and the lowest from molasses. Among the various forms of rice tested, best yield was obtained from undermilled black glutinous rice.

TABLE 1
THE CHEMICAL COMPOSITION OF RAW SUBSTRATES

Substrate	Moisture content (%)	Ash content (%)	Starch (%)	Reducing sugar as glucose (%)	Total N_2 (%)	Fat (%)
Cassava	10.7	2.8	66.9	0.95	0.56	0.81
Sorghum	12.3	2.9	63.0	0.13	1.49	1.75
Corn	12.1	1.3	63.7	0.22	1.58	0.34
Undermilled rice	12.3	3.1	66.9	0.11	1.32	2.25
Undermilled white glutinous rice	13.0	2.3	68.5	0.06	1.36	1.54
Undermilled black glutinous rice	12.6	1.3	66.9	0.15	2.10	2.68
Broken rice	12.4	0.5	71.8	0.23	1.06	0.42
Broken white glutinous rice	11.8	0.5	70.0	0.11	1.20	1.50
Molasses	-	-	-	40.9 (after hydrolysis)	0.46	0.08

TABLE 2
THE AMOUNTS OF ENZYMES USED IN CONVERSION OF VARIOUS SUBSTRATES

Substrate	Liquefying enzyme (Maxamyl) (g)	Conversion enzyme (Maxydrase) (g)
Cassava	0.0406	0.5066
Broken rice	0.0406	0.5066
Broken white glutinous rice	0.0406	0.5066
Corn	0.0812	0.5066
Sorghum	0.0812	0.5066
Undermilled rice	0.0812	0.5066
Undermilled white glutinous rice	0.0812	0.5066
Undermilled black glutinous rice	0.0812	0.5066

TABLE 3
PERCENTAGE OF CONVERSION

Substrate	Starch in flour (% w/w)	Whole volume of mash (ml)	Initial starch in mash (% w/v)	Final starch in converted mash (% w/v)	Conversion based on starch (%)	Glucose in converted mash (% w/v)	Actual reducing sugar derived from starch (% w/v)	Conversion based on glucose (%)
Cassava	66.9	1050	16.0	nil	100.0	17.3	17.1	95.9
Sorghum	63.0	1080	14.8	1.02	93.1	16.3	16.3	99.3
Corn	63.7	1100	14.7	1.67	88.6	15.4	15.4	94.4
Undermilled rice	66.9	1100	15.4	0.18	98.8	15.8	15.8	92.2
Undermilled white glutinous rice	68.5	1090	15.9	0.12	99.3	17.6	17.6	99.6
Undermilled black glutinous rice	66.9	1080	15.7	nil	100.0	17.4	17.4	99.7
Broken rice	71.3	1100	16.4	0.40	97.6	18.2	18.2	99.6
Broken white glutinous rice	70.0	1080	16.4	0.07	99.6	17.9	17.9	98.2

TABLE 4
COMPOSITION OF MASHES BEFORE AND AFTER FERMENTATION

Substrate	Before fermentation		After fermentation		
	Titrable acidity	Total reducing sugar as glucose (%)	pH	Titrable acidity	Final reducing sugar as glucose (% w/v)
Cassava	2.1	12.7	4.4	4.0	0.44
Sorghum	3.8	10.8	4.8	4.5	0.46
Corn	4.4	10.4	4.7	5.0	0.31
Undermilled rice	4.0	12.4	4.7	4.1	0.21
Undermilled white glutinous rice	4.7	11.2	4.5	4.7	0.30
Undermilled black glutinous rice	4.5	12.3	4.8	5.0	0.36
Broken rice	3.5	12.2	4.5	4.8	0.38
Broken white glutinous rice	4.5	13.8	4.6	4.9	0.35
Molasses	9.1	12.6	4.9	9.7	2.6

TABLE 5
YIELDS OF ALCOHOL

Substrate	Alcohol in distillate	Number of Proof gallon (100^o) per bushel of raw substrate		Actual yield of absolute alcohol obtained from raw substrate
		wet basis	dry basis	
	(% v/v)	(gal/bu)	(gal/bu)	(% w/w)
Cassava	11.1	4.68	5.2	30.2
Sorghum	10.0	4.20	4.8	27.1
Corn	9.3	3.90	4.4	25.1
Undermilled rice	10.0	4.20	4.8	27.1
Undermilled white glutinous rice	10.0	4.20	4.8	27.1
Undermilled black glutinous rice	11.0	4.64	5.3	29.9
Broken rice	10.1	4.25	4.9	27.4
Broken white glutinous rice	10.3	4.33	4.9	27.9
Molasses	10.3	4.33	---	13.5

Table 6 gives the fermentation efficiency obtained with various substrates. Under our experimental conditions, molasses was most efficiently utilized. Next in the order of efficiency come undermilled black glutinous rice, raw cassava, and sorghum respectively. The efficiency of fermentation obtained with corn and other forms of rice were practically the same, from about 67-71% if based on starch and 71-82% if based on the amount of sugar fermented.

Table 7 compares the cost of raw substrates yielding 1 kg of absolute alcohol.

DISCUSSION

The choice of raw materials for alcohol production depends on many things such as the price, the content of fermentable substances, the need for and the ease of conversion into fermentable sugars, the efficiency of fermentation, the cost per unit amount of alcohol produced, and finally the quality or grade (beverage or industrial) of alcohol desired.

For non-sugar raw materials, broken ordinary rice and broken glutinous rice have the highest starch content (Table 1), and they can be very effectively converted into fermentable sugars by the enzymes used (Table 3). The undermilled rices have but slightly less starch content than the broken rices and are quite as easily converted (especially the undermilled glutinous rice). The undermilled glutinous rices have higher fermentation efficiency than undermilled ordinary rice. The fermentation efficiency of the broken rices are about the same and as high as that of the undermilled white glutinous rice. The fermentation efficiency of undermilled black glutinous rice approachs that of the molasses. However the prices of the undermilled rices almost double those of the broken rices (Table 7), and large-scale production of alcohol from them will not be economical.

Traditionally, beverage alcohol made from rice is made exclusively from glutinous rice, be it the Thai rice beers (u and nam-khao) or the Chinese pae pow (rice wine). In the present investigation it was found that ordinary rice could be used with advantage. The cost of raw substrate yielding 1 kg of absolute alcohol was ฿ 0.17 less with broken

TABLE 6
FERMENTATION EFFICIENCY

Substrate	Theoretical yield of abs. alcohol based on starch (% w/w)	Theoretical yield of abs. alcohol based on sugar fermented (% w/w)	Actual yield of abs. alcohol[1] (% w/w)	Fermentation efficiency based on starch (%)	Fermentation efficiency based on sugar fermented (%)
Cassava	38.0	37.1	30.2	79.4	81.4
Sorghum	35.8	32.5	27.1	75.7	83.4
Corn	36.2	30.7	25.1	69.4	81.7
Undermilled rice	38.0	37.7	27.1	71.2	71.8
Undermilled white glutinous rice	38.9	34.6	27.1	69.6	78.3
Undermilled black glutinous rice	38.0	34.7	29.9	78.7	86.1
Broken rice	40.5	34.2	27.4	67.6	80.0
Broken white glutinous rice	39.7	36.1	27.9	70.3	77.5
Molasses	----	15.4	13.5	----	87.6

[1] From Table 5.

TABLE 7
COST OF RAW SUBSTRATE YIELDING 1 KG ABSOLUTE ALCOHOL

Substrate	Unit price (baht/kg)	Cost of raw substrate yielding 1 kg absolute alcohol (baht)
Dry cassava	0.73	2.42
Sorghum	0.67	2.47
Broken rice	0.76	2.78
Broken glutinous rice	0.83	2.97
Molasses	0.41	3.04
Undermilled ordinary rice	1.42	5.24
Corn	1.40	5.57

Note. Unit prices are the average for March 1972; source: ESG, ASRCT.

ordinary rice than with broken glutinous rice. The aroma of alcohol made from ordinary rice might not be the same as that from glutinous rice, but it is still good.

Alcohol made from black glutinous rice has a very pleasant aroma, peculiar to itself, but broken black glutinous rice is not available in the market. For a special blend of rice wine it might have some commercial value, if broken black glutinous rice could be obtained in big quantities and at reasonable price.

Sun-dried raw cassava has about the same starch content as those of the undermilled rices, but it is a little less readily convertible by the enzymes in term of the percentage of conversion, even though 100% conversion rate based on starch was obtained. This is because the enzymes, for some reason, did not quite completely break down cassava starch into glucose. The price of dry cassava is a little less than that of the broken rice, and its fermentation efficiency is quite high; hence cassava has the least raw material cost per unit amount of alcohol produced, compared with the other substrates tested, including molasses. The aroma of alcohol produced from cassava, though less pleasant than the alcohol produced from the other non-sugar substrate, is still better than that of alcohol made from molasses. It should be thought of when molasses is in short supply.

Sorghum and corn have the lowest starch content (around 63%), but sorghum is more readily convertible by the enzymes than corn is. The fermentation efficiencies of both substrates are quite high. The cost of raw sorghum yielding one kilogramme of absolute alcohol is next to the lowest, being only ฿ 0.05 less than that of cassava. However alcohol made from sorghum has a better aroma than that made from cassava. It has a special flavour(kaoliang wine) and is very popular among the Chinese.

Although corn is a popular substrate for beverage alcohol, especially in the U.S. (Kentucky bourbon), its per kilogramme price (1.40 baht) is prohibitive compared with the other non-sugar raw substrates.

Molasses is the cheapest raw material in our series so far as per kilogramme price is concerned, but molasses contains roughly only 40% of reducing sugar expressed as glucose. This is why raw material cost per unit amount of alcohol produced is rather high, or the highest of all the substrates except undermilled ordinary rice and corn, in spite of the fact that molasses is the most efficiently fermented of all substrates tested. However, molasses has one distinct advantage in that it can be utilized by the yeast without having to be converted nor does it need any elaborate pre-treatment. For the production of industrial alcohol by the existing distilleries in the country, it is still the substrate of choice.

Conversion of starch to fermentable sugars by commercial enzymes is not practiced by distilleries in this country. The Bang Yi Khan Distillery (Government-owned, but leased to Mahakhun Distillery Co., Ltd.) uses mold bran prepared at the distillery to convert steamed broken white glutinous rice to produce a special brand of rice spirit (the so called glutinous rice wine or pae pow), but the conversion rate is rather low. The effectiveness of commercial enzymes used in the present study (Table 3) is very satisfactory in all cases, but larger amounts of enzymes than those recommended by the producer had to be used. This is because the enzymes have been kept in our laboratory (4°C in a refrigerator) for about 4 years. When freshly received the recommended dosage worked all right, but now four times as much Maxamyl were required for liquefying cassava and the broken rices, while eight times as much were required for sorghum, corn, and the undermilled rices (Table 2). Twice as much Maxydrase was required for conversion of all the substrates tested.

To use commercial enzymes in starch conversion for alcohol production would require additional costs. A section for starch conversion has to be set up in the plant. This would incur a certain amount of new expenditure. The process cost would have to be worked out also. How much this would all add up to needs further study. Cost comparison between enzymatic conversion and acid hydrolysis also has to be made before a definite conclusion could be drawn.

In order to develop and expand the distilling industry, a distinction between beverage alcohol and industrial alcohol should be made. Markets for special types of beverage alcohol have to be developed both at home and abroad. New uses of alcohol, such as mixing it with gasoline for use as automobile fuel, would certainly increase the demand for industrial alcohol. If export of industrial alcohol is aimed at, the management of local distilleries will have to be improved. Full use of manpower and existing equipment must be accomplished and wastage reduced to a minimum. The production cost could then certainly be drastically cut. Unless these conditions are corrected, the chance of exporting industrial alcohol will be as remote as ever.

CONCLUSION

1. Conversion of starch to fermentable sugars by commercial enzymes for alcohol production was found to be effective, and the costs of conversion by this method should be further studied.

2. For the production of industrial alcohol under existing conditions in local distilleries, molasses is still the substrate of choice. Dry cassava chips with the proper method of conversion could substitute for molasses when the latter is in short supply.

3. Sorghum and broken rices appear to be the substrates of choice for beverage alcohol, but an economical method for starch conversion has to be worked out.

4. Corn and undermilled rice were found to be far too expensive for alcohol production.

5. Alcohol made from black glutinous rice has a special aroma peculiar to itself. This substrate might be useful for the production of a unique type of rice wine.

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REFERENCES

HORWITZ, W., ed. (1965).—"Official Methods of Analysis of the Association of Official Agricultural Chemists." (The Association of Official Agricultural Chemists: Washington, D.C.)

STARK, W.H., ADAMS, S.L., SCALF, R.E., and KOLACHOV, P. (1943).—Laboratory cooking, mashing, and fermentation procedures. Ind.Engng Chem.analyt.Edn 15 (7): 443-446.