Functional Properties of Brewers' Liquid Adjuncts[1]

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ABSTRACT

Enzyme technologies that may be applied to produce varied carbohydrate profiles are reviewed, with emphasis on the role of maltose in fermentation. Pertinent manufacturing equipment is considered. Brewing quality of corn syrup is examined, and differences between acid-hydrolyzed and enzyme-hydrolyzed starches are noted. Reference is made to some of the unfermentable sugars in commercial corn syrup.

Key words: *Bitter sugars, Carbohydrate profile, Flocculation, High maltose, Jet cooking, Unfermentable sugars*

Although corn wet millers had developed some interest in the preparation of liquid adjunct for brewing as early as 1933, serious investigation was not initiated until after World War II. The product was considered as a total or partial replacement for the corn-grits extract.

Product design (carbohydrate profile) was regulated to a great extent by the technology then existing. As technology progressed, more sophisticated products were produced. Improved analytical techniques provided means to evaluate and distinguish unique functional properties.

Brewers' liquid adjuncts can have a significant impact on the brewer and his product. A better understanding of the properties by which an adjunct influences the aforementioned factors and the control or modification of those properties may aid in the selection of adjuncts.

Many characteristics of liquid adjuncts influence properties in beer and beermaking, including flavor, rate and degree of fermentation, yeast propagation, filterability, and clarity.

Flavor

Acid hydrolysis of grain products is considered to modify fatty and protein constituents, resulting in off-flavored materials. Acid hydrolysis contributes to the production of miscellaneous sugar products that, in turn, can contribute to variable flavor and fermentability.

Ough (8) reported on the analysis of 11 glucodisaccharides present in acid-hydrolyzed corn syrup (60-dextrose equivalent). Although maltose was the predominant disaccharide generated, up to 20.9% of the disaccharides reported was identified as nonmaltose.

Fetzer et al (6) examined acid-thinned corn syrup and observed that the acid-heat treatment caused increased color and bitterness.

Taylor and Lifschitz (10) heat treated starch, corn amylose, and dextrose under the conditions of acidity and temperature employed in the commercial hydrolysis for corn sugar. The reaction with dextrose produced 1.5% gentiobiose. Gentiobiose is an unfermentable disaccharide that has a bitter flavor. The study (10) indicated that disaccharides are produced by reversion, ie, repolymerization. The other carbohydrates produced negligible bitterness. Gentiobiose was concluded to arise from direct scission of starch. A further conclusion is that heat and acidic conditions promote the formation of gentiobiose, which contributes to off flavors.

Fermentability

The composition of a liquid adjunct may affect primary fermentation rate and degree (Real Degree of Attenuation).

Commercial brewers' corn syrups are prepared by an acid-enzyme process. The carbohydrate is subjected to a thinning process that employs high temperatures and acidic conditions. The acidity of the resultant carbohydrate is neutralized and then further converted, using microbial enzymes. Typical syrups contain 35% monosaccharides and 35% disaccharides.

Masschelein et al (7) reported that yeasts grown on a dextrose medium inadequately developed maltozymase, depreciating the fermentation rate of maltose. Akin and Krabbe (1) concluded that worts with a monosaccharide content higher than 10% on a total carbohydrate basis required extended periods for end fermentation. Witt and Blythe (11) investigated the fermentability of malt worts supplemented with 35% acid-thinned and 35% enzyme-thinned corn-syrup solids, respectively. Under pilot brewing conditions, the worts containing the acid-thinned syrup showed a slower fermentation rate, indicating about 3% lower Real Degree of Attenuation after 192 hr at 12–13°C. This difference was accentuated with comparable high-gravity wort.

The bitter flavor attributed to the disaccharide gentiobiose has been noted. Other disaccharides generated, particularly isomaltose, are unfermentable and consequently reduce the degree of fermentability.

Filterability and Clarity

Clarity can be effected by liquid adjuncts. As reported by Akin and Krabbe (1), worts containing more than 10% dextrose on a total carbohydrate basis required extended periods for fermentation. Cooling the beer before exhaustion of fermentable sugars prevented flocculation.

EXPERIMENTAL

Brewing

Procedures, equipment, and pertinent fermentation practices were conducted as outlined by Witt and Blythe (11).

Primary Beer Filtration

Single-plate Sparkler Filter (9-in. diameter), constant CO_2 head pressure (5 psi), and diatomaceous earth precoat ("Hi-Flow," Manville Corp.) were used.

[1]Presented at the 47th Annual Meeting, Miami, FL, May 1981.

Viscosity Determinations

Brookfield. Viscosities of mash, corn, or starch suspension were determined using Brookfield viscometer R.V.T.; results are reported in centipoises (cp).

Brabender. The mechanics of Brabender Amylograph performance were given by Elder and Schoch (5). From these data, it may be calculated that 1.0 Brabender unit is approximately equivalent to 1.75 cp.

Carbohydrate Profile

Liquid chromatography and gas chromatography, were used to determine the wort, corn syrup, and beer profiles.

Liquid Chromatography. Mono-, di-, and trisaccharides were resolved by liquid chromatography (3/8 × 18 in. column, Aminex 50W—X4, cation exchange, calcium form) at 85°C. The basic procedure used is outlined by Brobst et al (4).

Gas Chromatography. Gas chromatography was used to determine isomaltose and maltose. Materials were diluted with water and freeze-dried. The "anhydrous" residue was dissolved in pyridine and treated with hexamethyldisilazane and trifluoroacetic acid to prepare the trimethylsilyl derivatives. The derivatives were resolved on a 1/8 × 8 in. column of OV-17 on Chromosorb W. A temperature program cycle ranged from 150 to 310°C, with disaccharides being eluted in the range of 190–230°C. Some details of this procedure were outlined by Brobst and Lott (3).

TABLE I
Fermentation of Variable Substrates

	Samples[a]			
	I	II	III	IV
Degrees Plato	12.9	13.5	14.7	14.2
Wort Carbohydrate Profiles (g/100 g liquid)				
Dextrose	0.963	1.032	2.499	0.562
Maltose	5.445	6.220	5.784	6.671
Isomaltose	0.00	ND[b]	0.196	0.00
Maltotriose	1.404	1.409	2.302	2.440
Higher	3.796	3.487	3.257	3.105
Beer Carbohydrate Profiles (g/100 g liquid)				
Dextrose	0.058	0.018	0.111	0.000
Maltose	0.144	0.152	0.332	0.081
Isomaltose	0.00	0.083	0.196	0.01
Maltotriose	0.679	0.551	1.214	0.902
Higher	3.646	3.365	3.189	2.789
Triose Depletion (%)	51.6	60.5	47.3	63.0
Estimated DA (%)[c]	63.2	67.4	71.2	69.2
Determined DA (%)[c]	63.1	63.9	66.0	67.7

[a] I (control) = 60% malt–40% corn grits wort. II = All-malt wort 8.1°P, acid-enzyme corn syrup added to 13.5°P (60% malt–40% corn syrup solids). III = Acid-enzyme corn syrup only carbohydrate source. IV = Enzyme-enzyme corn syrup only carbohydrate source.
[b] ND = None detected.
[c] Degree of attenuation.

$$\text{Estimated DA (\%)} \frac{\text{mono + di + tri in wort, g/100 ml}}{\text{total carbohydrate in wort, g/100 ml}} \times 100$$

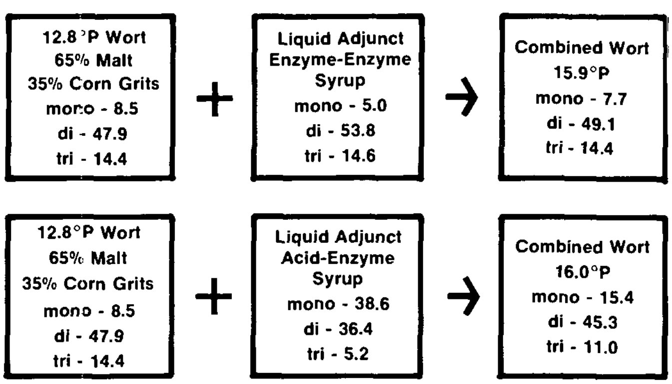

Fig. 1. Wort carbohydrates as affected by addition of corn syrups. Carbohydrate profiles expressed as percent carbohydrate. Malt (52.3%) + corn grits (28.1%) + corn syrup solids (19.6%) = 100% extract.

RESULTS AND DISCUSSION

Effect of Preparation on Substrate Composition and Performance

Worts that contained different carbohydrate substrates were prepared. Carbohydrate profiles were made on the samples before and after fermentation; pertinent data are given in Table I. To ensure proper fermentation conditions, nutrients were added to samples III and IV as described by Witt (12). The results illustrate the following factors.

As indicated by the content in beers II and III, acid conversion promotes an increase in isomaltose. It is particularly evident in substrate III that acid conversion results in an increase in the ratio of dextrose to maltose. The beer prepared with all acid-enzyme substrate (III) demonstrated reduced triose depletion.

In contrast to substrates containing acid-enzyme syrups, substrates of enzyme-enzyme syrups showed a reduced dextrose to

TABLE II
Yeast Population[a]

Time of Fermentation (days)	Enzyme-Enzyme (Cells × 10^6/ml)	Acid-Enzyme (Cells × 10^6)
1	56.8	42.3
2	58.2	53.6
3	47.8	45.0
4	47.8	22.5
7	18.4	19.2
8	16.0	17.0

[a] The wort was 72 in. deep, and samples were collected at a depth of 20 in. Cells × 10^6.

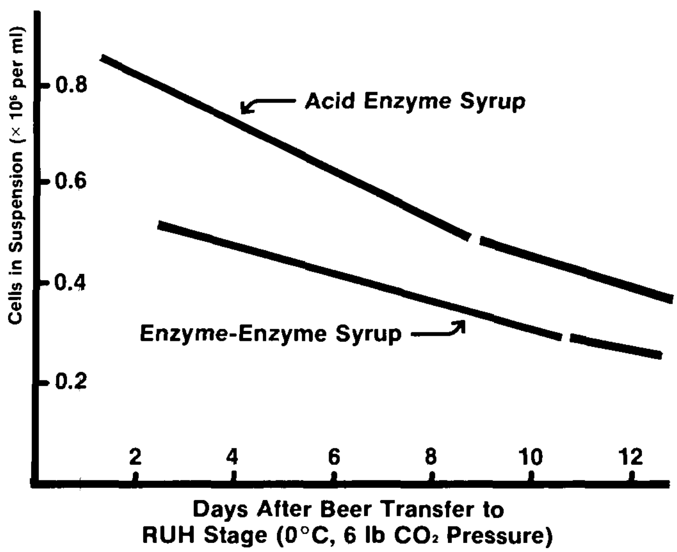

Fig. 2. A comparison of suspended yeast cell counts of brews prepared with enzyme-enzyme and acid-enzyme converted syrups.

maltose ratio; contained less isomaltose; and demonstrated increased triose depletion.

The determined Degree of Attenuation (DA) and the estimated Degree of Attenuation (Estimated DA) agree quite closely for the control and the enzyme-enzyme syrup beers. In contrast, the acid-enzyme syrup beers resulted in a relatively reduced attenuation, by greater than 4%.

The determined Degree of Attenuation is the percent reduction in carbohydrate due to fermentation (2). Estimated Degree of Attenuation is an arbitrary value, arrived at by experience, which corresponds to the sum of monosaccharides plus disaccharides plus 0.66 times trisaccharides, expressed as a percentage of the total measured carbohydrates. For example, substrate I was calculated as follows: mono (0.963 g) + di (5.445 g) + (0.66) tri (0.927 g) = 7.335 g estimated fermentable carbohydrate. Total carbohydrate is 11,608 g. Estimated Degree of Attenuation is 63.2%, as estimated by dividing 7.335 g (100%) by 11.608 g.

Effect of Dextrose Content

Another study was conducted to demonstrate the effect of dextrose in the liquid adjunct. Data from that study (Fig. 1) represent grain wort (malt-corn grits) supplemented to higher solids with enzyme-enzyme and acid-enzyme corn syrups, respectively. The acid-thinned corn syrup is the more conventional brewers' liquid adjunct. During fermentation, samples were tested for yeast cell population; the results are given in Table II. The yeast population during the seventh and eighth days is similar; the somewhat higher number in the acid-enzyme wort is indicative of the greater concentration of suspended yeast during the RUH (aging) period.

After transfer to the RUH stage, the beer was monitored for suspended yeast cells. Figure 2 illustrates the relative change. The beer prepared with the enzyme-enzyme syrup consistently displayed a lower yeast count, coinciding with improved flocculation.

Clarity was also monitored during the RUH stage; the absorbance was measured at 700 nm (Fig. 3). The beer prepared with enzyme-enzyme corn syrup again demonstrated improved flocculation, as indicated by a lower absorbance.

A further indication of flocculating properties is the increased time required for primary filtration. Although the initial filtration time for each was the same (7 min for the first 5 L), the beer prepared with acid-enzyme corn syrup required nearly twice as long (135 vs 75 min) as that prepared with enzyme-enzyme corn syrup.

The results of this study confirm that a high dextrose content can adversely affect fermentation, yeast flocculation, residual yeast concentration, and related filterability.

Effect of Adjunct on Economics

We have placed little emphasis on the effect that liquid brewers' adjuncts may have on the economics of brewing. Brewers are well aware that economics are significantly affected by properties such as degree and rate of attenuation, the ability to ferment high-gravity wort efficiently, and the achievement of good filtration properties.

Economics can be improved by on-site preparation of liquid adjuncts by permitting the brewer to use many of the substrates at his disposal (eg, corn flour, corn grits, rice, wheat, rye, and starch).

It is important that the qualities of the adjunct be retained while maintaining a practical process for their preparation. Consideration of the cost of material and equipment, and of energy consumption must also be of concern if on-site utilization of substrates is undertaken.

Conversion Process

We have stated that an acid-conversion process adversely contributes to the properties of a brewers' liquid adjunct. This, then, leads to the examination of a "total" enzyme conversion process.

Conventional Batch. Conventional batch enzyme-conversion procedures have limited practical applications. During gelatinization, viscosity increases dramatically, making it difficult to handle high solids. High energy loads are required to achieve suitable mixing. Enzyme action is not uniform and is inefficient, as illustrated in Fig. 4[2]. Viscosity increases significantly as starch (carbohydrate) is gelatinized. When a conventional bacterial α-amylase is employed, the peak viscosity is reduced with relatively high final viscosity. When a thermally stable bacterial α-amylase is employed at a comparable dosage level (percent weight on dry solids), a higher peak viscosity is encountered with a lower final viscosity. A blend of enzymes reduces the peak viscosity and final

[2]Data provided by Novo Laboratories, Inc.

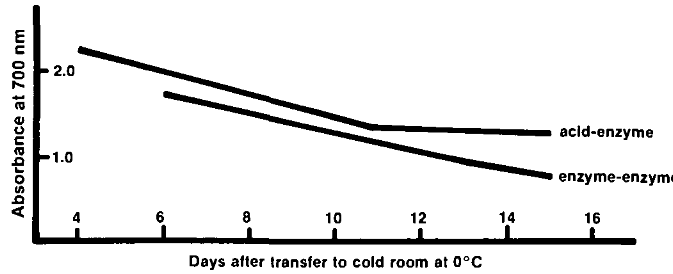

Fig. 3. A comparison of clarity of brews prepared with enzyme-enzyme and acid-enzyme converted syrups.

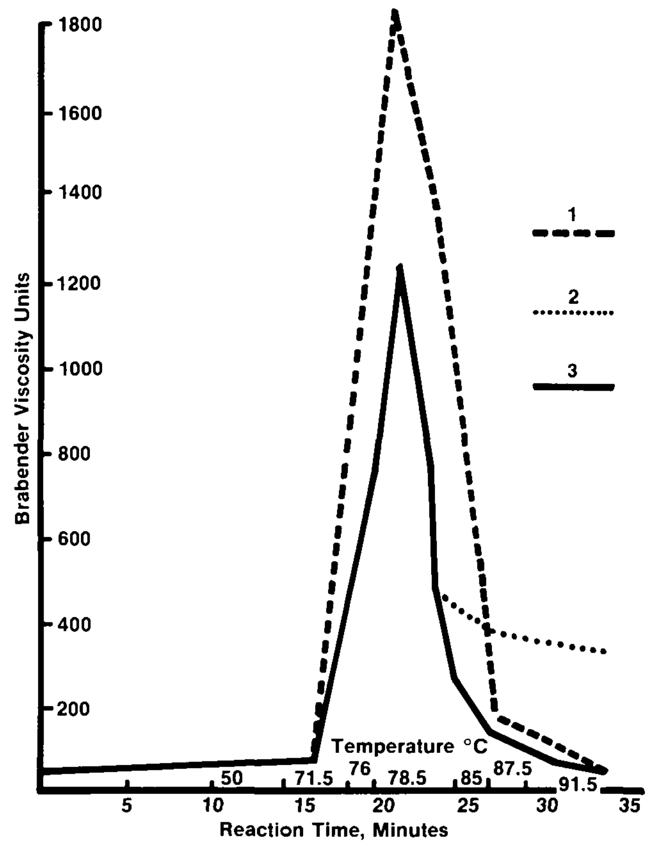

Fig. 4. Viscosity characteristics of batch enzyme conversions. Curve 1 = 28% starch solids and 0.12% thermally stable bacterial α-amylase. Curve 2 = 0.12% conventional bacterial α-amylase. Curve 3 = 0.12% mixture of 50% thermally stable bacterial α-amylase and 50% conventional bacterial α-amylase.

viscosity.

These differences result primarily from two factors: a difference of enzyme activity, and a difference of temperature optima of enzymes. The differences provide insight into carbohydrate conversion properties, particularly demonstrating that starch is not completely gelatinized at 80°C (176°F).

Data comparing pertinent enzyme values are given in Table III.

Another experiment (Fig. 5) further demonstrated incomplete gelatinization at 90°C by noting a continual rise in viscosity. Additionally, the limited utility of malt as an α-amylase source in batch conversion is illustrated.

Jet Cooking. Many of the difficulties encountered in batch conversion can be eliminated or minimized by jet cooking. Figure 6 illustrates a process employed.

Substrate is prepared in the slurry tank at the desired solids concentration and pH and may contain desirable additives such as calcium for enzyme stabilization. The substrate pumped through a jet is instantaneously gelatinized by live steam injection heating to 300°F or higher. The material is thoroughly dispersed by maintaining that temperature in a coil for a period of 2 min or longer. The gelatinized product is reduced to atmospheric pressure and temperature in the flash tank. The substrate is discharged from the flash tank into a reaction vessel in which the desired enzyme system is employed.

Jet cooking provides a thoroughly gelatinized paste, which is an ideal substrate for enzyme conversion. Peak viscosities are eliminated, and energy demands reduced.

The utility of the enzymes of malt (a natural ingredient) is enhanced by the jet-cooking process. A patent (13) has been issued describing the application of malt enzymes to jet-cooked starch.

An example of the application of malt infusion enzyme is presented in Fig. 7. Enzyme activity is expressed in SKB 30°C units

TABLE III
Relative Comparison of Amylase Enzymes

Curve[a]	Enzyme	Relative Activity
1	0.12% thermally stable bacterial α-amylase	×2
2	0.12% conventional bacterial α-amylase	×1
3	0.06% conventional bacterial α-amylase 0.06% thermally stable bacterial α-amylase	×1.5

[a] Refers to curve in Fig. 4.

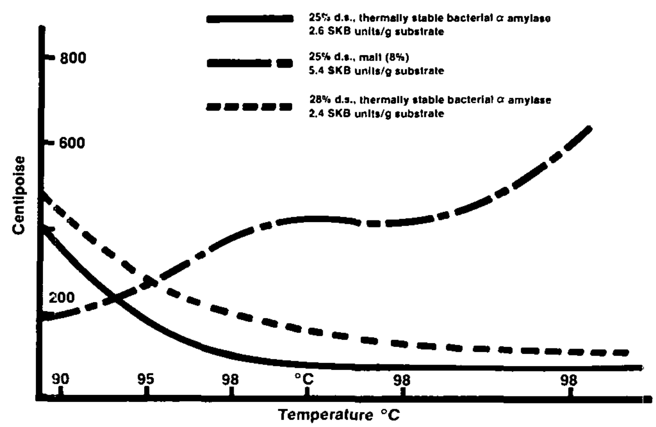

Fig. 5. Viscosity characteristics of batch-converted corn grits. Malt vs thermally stable bacterial α-amylase. ——— = 25% d.s., thermally stable bacterial α-amylase giving 2.6 SKB units per gram of substrate. —— - —— = 25% d.s., malt (8%) giving 5.4 SKB units per gram of substrate. - - - - = 28% d.s., thermally stable bacterial α-amylase giving 2.4 SKB units per gram of substrate.

(9). Viscosity is rapidly reduced to a level that is easily handled, and is saccharified to prepare a liquid adjunct.

The liquefied product, whether from a conventional batch or jet-cook conversion, can be saccharified utilizing numerous techniques and enzyme combinations to achieve specific goals.

The data in Fig. 8 present several alternatives. Cornstarch was liquefied at 30% dry solids, using the jet-cooking process described and liquefaction with bacterial α-amylase. The product (cycle I) containing the described carbohydrate composition was saccharified with malt infusion as a source of α-amylase. The high-maltose product obtained would be well suited for application as a liquid adjunct of estimated 69.2% fermentability.

The above product further treated with an enzyme blend of pullulanase and glucoamylase appreciably increased the dextrose content with negligible change in the maltose content, increasing the fermentables to 85.7%.

In Cycle II (Fig. 8), the liquefied product was saccharified with a blend of malt infusion as a source of α-amylase and pullulanase directly, resulting in a high-maltose adjunct that is 81.1% fermentable.

Liquefied product in Cycle III (Fig. 8) was prepared similarly to I

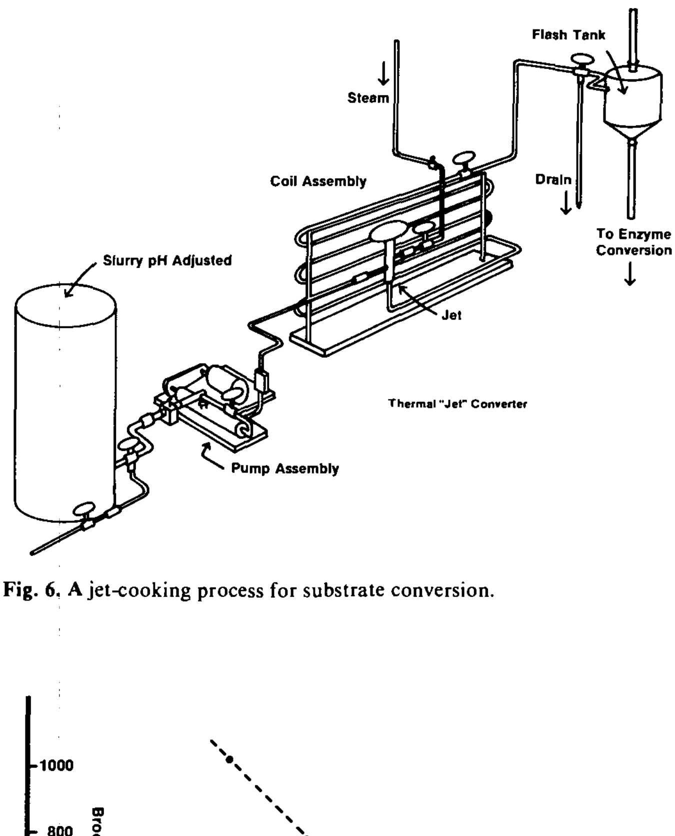

Fig. 6. A jet-cooking process for substrate conversion.

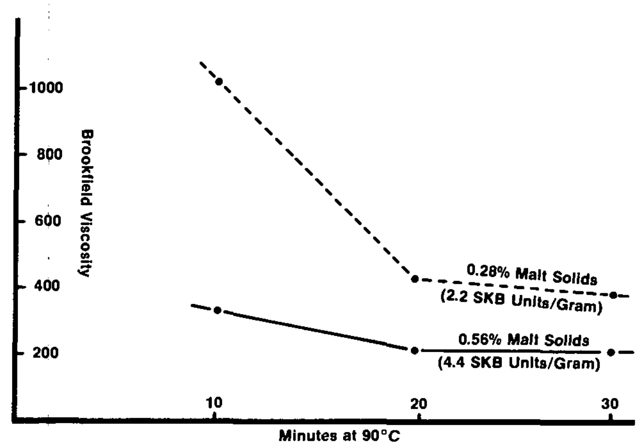

Fig. 7. Liquefaction of 40% jet cooker starch at 90°C by malt infusion (pH 6.5; 0.04% Ca++ in starch).

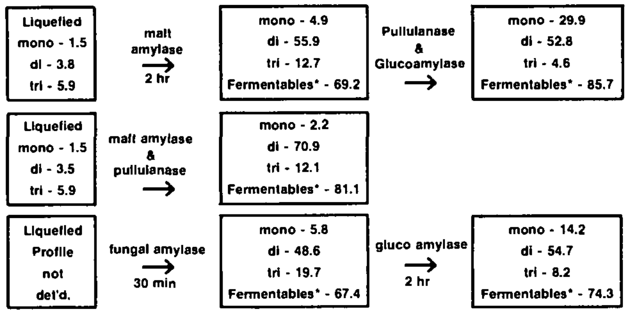

Fig. 8. Conversion of enzyme-liquified starch with 30% dry solids (pH 5.0, 62°C). Syrup profiles expressed as percent carbohydrate. Fermentables – estimated = mono + di + 0.66 tri.

and II, but the carbohydrate profile was not determined. It was saccharified for 30 min with a fungal α-amylase, resulting in a high-maltose liquid adjunct containing 67.4% fermentables. Further treatment with a very low level of glucoamylase again gave a significant increase in dextrose, without sacrificing maltose content, and an increase in total fermentables to 74.3%.

The data in Fig. 8 illustrate the many alternatives available to the brewer for on-site preparation of a liquid brewers' adjunct. Because of its FDA status, pullulanase may be unsuitable; however, the products can be prepared with fully acceptable enzymes such as bacterial α-amylases, fungal amylases, glucoamylases, and malt amylases.

CONCLUSION

Several factors influence the decision when considering selection of, or preparation methods for, brewers' liquid adjunct. Included are cost, handling, rate and degree of attenuation, labeling, and resultant beer quality (clarity and flavor). Selecting the most beneficial brewers' liquid adjunct is a challenge. However, with the aid of new processing techniques and more sophisticated analytical procedures, the brewer can develop a more comprehensive background that will allow facing those challenges with confidence.

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