Annex 9-8

Sugar Syrup (DE 50–70) from Corn Flour

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Starch in corn flour (22% solids, (w/w) was hydrolyzed by different concentrations of α-amylase and 12 KNU/100g Suspension produced a sugar syrup with dextrose equivalent (DE) of 58%. As tap water contained calcium, addition of calcium acetate did not improve starch hydrolysis. To increase total solids, either corn flour or corn flour suspension was added during liquefaction. Among the different conditions studied, addition of corn flour suspension (30g of 33%, w/w) to liquefying starch (22%, w/w) produced a sugar syrup with highest DE (DE 60) at 3.0 h. To increase the DE, malt extract rich in β-amylase was added to liquefied starch preparations supplemented with either corn flour (10 g) or corn flour suspension (33%, w/w) and DE increased from 50 and 60 to 62.2 in both, respectively. Then malt powder suspension (25%, w/w) equivalent to malt extract was added directly and DE obtained were 57.0 and 55.21, respectively. These results indicate that glucose syrups of DE 50 and 70 can be prepared using α-amylase alone without malt β-amylase.

1 Introduction

Sugar syrups (DE 50–70, high conversion syrups [1]) obtained by starch hydrolysis have increased contents of maltose with reduced amounts of higher sugars [2]. Thus they have reduced viscosity. In addition they show increased moisture holding capacity [3] and higher fermentability. Sugar syrups of DE 50–55 are preferred in hard confectioneries, brewing and fermentation industries [1, 4], while sugar syrups of DE 56–70 are utilized in confectioneries (soft), soft drinks, brewing and fermentation industries and in the preparation of jams, conserves and sauces [4]. High conversion syrups are produced by saccharifying liquefied starch with β-amylase or fungal α-amylase and glucoamylase [1, 3, 5]. In this paper we report the preparation of sugar syrup of DE 50–70 using starch in corn flour as raw material. To increase the total dry substance in sugar syrup, hydrolysate was supplemented with corn flour during hydrolysis. In addition, effect of malt amylase was determined.

2 Experimental

2.1 Materials

α-Amylase (Termamyl® 60L, activity 67.5 KNU g^{-1}) was from NOVO Industries (Denmark). Corn was purchased from local market and pulverized in a domestic grinder.

2.2 Analytical methods

Reducing sugar produced by hydrolysis of starch in corn flour was determined by 3,5 dinitrosalicylic acid method and

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represented in terms of glucose [6]. Total and reducing sugars [7], and dry substances in extract [8] and recovery of starch [8] in terms of reducing sugar were determined [8]. The DE of sugar syrup was calculated as described elsewhere [2].

2.3 Preparation of malt powder and malt extract

Malted corn [9] (1000 g) was dried at 40 °C for two days and powdered by a domestic grinder. Malt powder suspension (25.0 %, w/w, at pH 4.0) in distilled water was mixed for 30 min and strained through a muslin cloth. Malt extract was taken as β-amylase source and the activity was determined [9].

2.4 Effect of a α-amylase concentration on the hydrolysis of starch in corn flour

Corn flour (22.0 %, w/w) in suspension (tap water, pH adjusted to 7.0) was hydrolyzed for 3.0 h by different concentrations of α-amylase (from 3.0 to 24.0 KNU/100g suspension) at 98 °C. Reducing sugar [6] and recovery of starch [8] were determined.

2.5 Effect of calcium on the hydrolysis of starch in corn flour

Corn flour (22.0 %, w/w) in suspension (tap water, pH 7.0) containing 0.05 g of calcium acetate (0.05 %, w/w) was hydrolyzed by optimized amount of α-amylase at 98 °C. To the control no calcium acetate was added.

2.6 Hydrolysis of corn flour while supplementing with corn flour/corn flour suspensions

Corn flour (22.0 %, w/w) in suspension (tap water, pH 7.0) was hydrolyzed at 98 °C. At 1.0 and 2.0 h, either each of 5.0 g corn flour or a suspension of 5.0 g corn flour in 10.0 ml tap water or a suspension of 10.0 g corn flour in 10.0 ml of tap water was added and continued the hydrolysis for another hour. This supplementation works out to a total addition of either 10.0 g of corn flour or a corn flour suspension of 30.0 g (33.0 %, w/w) or 40.0 g (66.0 %, w/w) respectively.

2.7 Hydrolysis of corn flour hydrolysate by malt amylase in malt extract/powder suspension

The pH of corn flour suspension (22.0 %, w/w) hydrolyzed while supplementing with either corn flour (10.0 g) or corn flour suspension (30.0 g of 33 %, w/w) was adjusted to 4.0. At 3.0 h either 33.0 ml of malt extract (extract of 25.0 %, w/w malt powder suspension) or 80 g of malt powder suspension (25.0 %, w/w) was added. The mixture was incubated for 3.0 h at 60 °C.

3 Results and Discussions

3.1 Effect of α-amylase concentration on the hydrolysis of starch in corn flour

Different α-amylase concentrations used for hydrolysis of 100.0 g corn flour Suspension (22 %, w/w) were from 3.0 to 24.0 KNU. This is equivalent to 7.33 and 0.91 g DS/KNU or 5.64 and 0.7 g starch/KNU, respectively (Fig. 1). Moisture and starch content of corn flour were 12.0 % and 77.0 %, respectively [8]. Hydrolysis by 12.0 and 24.0 KNU/100 g suspension gave similar results (Fig. 1). At 3.0 h, 95.7 and 93.0 % of the added starch was respectively recovered as sugar/dextrins when 12.0 and 24.0 KNU/100 g suspension were used (Table 1). Dry substance in the extract was 12.8 %

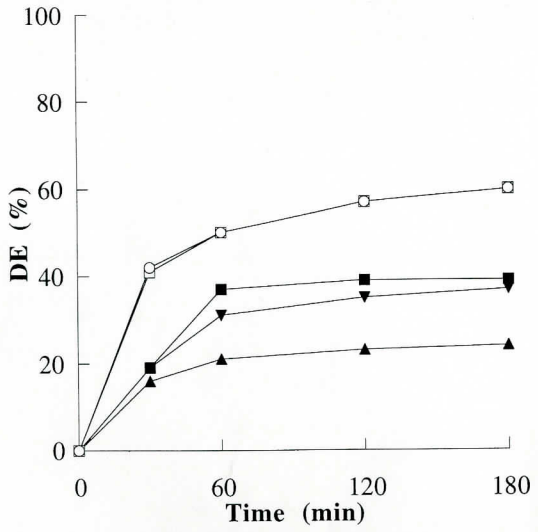

Fig. 1. Effect of different concentrations of α-amylase activity on the hydrolysis of starch in corn flour as a function of time at pH 7.0 and 98 °C.
(▲), 3.0; (▼), 6.0; (■) 9.0; (❑), 12.0; (●), 18.0 and (○), 24.0 KNU/100 g corn flour suspension.

Table 1. The dextrose equivalent (DE) and dry substance (DS) obtained and the starch recovered as reducing sugars in the extract of corn flour hydrolysate while using 12.0 and 24.0 KNU α-amylase per 100 g of corn flour suspension (22 %, w/w) at 95 °C and pH 7.0.

Parameters	α-Amylase/100 g corn flour suspension	
	12.0 KNU	24.0 KNU
DE (%)	60.0	60.0
DS (g)	12.8	13.5
Recovery (%)	95.7	93.0

and 13.5 %, respectively (Table 1). DE formed at different time intervals by the hydrolysis of the above two different concentrations of α-amylase were almost same (Fig. 1) and hence α-amylase activity of 12.0 KNU/100 g suspension of corn flour (22 %, w/w) was selected. A continuous liquefaction of starch was reported by *Carr* et al. [10]. In this process they have used 0.0–4.0 % α-amylase to hydrolyze 50.0–60.0 % starch at 98 °C. In our experiment the optimum enzyme concentration used was 0.2 % to hydrolyze 16.9 % starch. Thus starch/enzyme ratio works out to be 84.5 when compared to 12.5 and 15.0 reported earlier [10].

3.2 Effect of calcium on the hydrolysis of starch in corn flour

DE obtained by the hydrolysis of corn flour suspension containing 0.05 % (w/w) calcium acetate was same as that obtained with suspension containing no calcium acetate (Fig. 2). Since calcium stabilizes α-amylase and increases its activity [11, 12, 13] it was added to the reaction mixture. The insignificant difference in DB could be due to high content of calcium (215 mg · l^{-1}) present in tap water [14]. Thus for corn flour suspension tap water could be used without of calcium acetate supplementation.

3.3 Hydrolysis of starch in corn flour while supplementing with corn flour/corn flour suspensions

As dry substance obtained in extract by the hydrolysis of 22.0 % (w/w) corn flour suspension was 12.8 % (Table 1), and it was difficult to mix when more corn flour was taken as

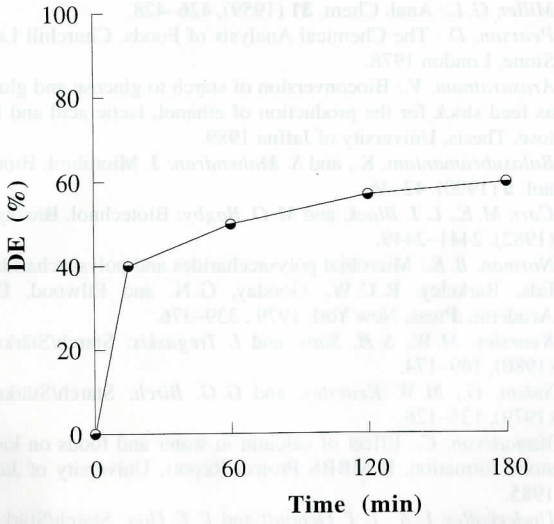

Fig. 2. Effect of calcium acetate on the activity of α-amylase at pH 7.0 and 98 °C.
(●), With and (○), without calcium acetate (0.05 %, w/w).

suspension in the initial stages, it was tried to add either corn flour or corn flour suspensions (33.0 %, w/w or 66.0 %, w/w) at two stages during the hydrolysis of 22.0 % (w/w) corn flour suspension. The results obtained at 3.0 h of hydrolysis after the addition of either corn flour or corn flour suspensions are given in Table 2. When either corn flour or corn flour suspensions were added to liquefied starch resulted from the hydrolysis of 22.0 % (w/w) corn flour suspension, reducing sugars obtained were higher in former condition than in conditions where no additional solids were added. Reducing sugar and DE obtained were highest when hydrolysate was supplemented with 33.0 % (w/w) corn flour suspension. However dry substance obtained was highest when hydrolysate was supplemented with corn flour (Table 2). Recovery was high when the hydrolysate was supplemented with either corn flour or corn flour suspension (33.0 %, w/w). Thus comparatively corn flour hydrolysate supplemented with either corn flour or corn flour sus pension (33.0%, w/w) seems to be promising. Hence these two methods were considered to increase the dry substance in extract of corn flour hydrolysate. However DE obtained did not exceed a value of 60.0. Thus it was decided to use β-amylase from malt extract anticipating that this plant origin [4, 5, 15] enzyme can act on amylopectin and result in 50.0–60.0 % conversion of starch in corn flour to maltose [5] leaving β-limit dextrins [12].

Table 2. Total and reducing sugars, DE and DS obtained in the extract and starch recovered by the hydrolysis of 22 % (w/w) corn flour suspension while supplementing with either corn flour or corn flour suspensions. The data given below were measured at 3.0 h.

Parameters	Supplemented with			
	Nil	Corn flour	Corn flour suspension (w/w)	
			33 %	66 %
Reducing sugar ($g\ l^{-1}$)	100.8	115.0	120.0	116.0
Total sugar ($g\ l^{-1}$)	180.0	230.0	200.0	320.0
DE (%)	56.0	50.0	60.0	36.0
DS (g)	12.8	26.1	20.8	22.8
Recovery (%)	95.7	93.0	83.0	45.0

Table 3. Total and reducing sugar, DE and DS in the extract obtained by the hydrolysis of 22.0 % (w/w) corn flour suspension while supplementing with either corn flour or corn flour suspension (33 %, w/w). Hydrolysis was carried out with α-amylase (12.0 KNU/100g initial corn flour suspension) and with β-amylase either in malt extract (33.0 ml) or in malt powder suspension (25.0 %, (w/w). The data given below were measured at 6.0 h (total hydrolysis time).

Parameters	Corn flour 10.0 g		Corn flour suspension (33.0 %, w/w)	
	Malt		Malt	
	Extract 33 ml	Suspension (25 %, w/w)	Extract 33 ml	Suspension (25 %, w/w)
Reducing sugar ($g\ l^{-1}$)	115.8	118.2	102.6	82.2
Total sugar ($g\ l^{-1}$)	193.0	190.0	180.0	149.0
DE (%)	62.2	57.0	62.2	55.2
DS (g)	27.0	16.0	23.0	15.0
Recovery (%)	78.0	4.0	77.0	57.0

3.4 Hydrolysis of corn flour hydrolysate by malt amylase in malt extract or malt powder suspension

Reducing sugars and dextrose equivalent obtained varied when corn flour hydrolysate obtained by hydrolysis of corn flour while supplementing with corn flour or corn flour suspension (33 %.0, w/w) and then by β-amylase in either malt extract or malt powder suspension (malt amylase activity was 0.43 μ mole glc./min/g malt powder) (Table 3). DS and DE of sugar syrup and recovery of starch were high when the hydrolysate was supplemented with corn flour and then hydrolyzed by β-amylase in malt extract (Table 3). Recovery of reducing sugars were almost same when the hydrolysate was supplemented either with corn flour or corn flour suspension (33.0 %, w/w) and then hydrolyzed by β-amylase in malt extract. Therefore, among the four different methods considered, hydrolysis of corn flour hydrolysate while supplementing with corn flour and then hydrolyzing with β-amylase in malt extract seems to be the best. However, if the best results obtained in this set of experiments (Table 3) are compared with previous set of experiments (Table 2) where corn flour hydrolysate was supplemented either with corn flour or corn flour suspensions (33 %, w/w) and hydrolyzed only by α-amylase, hydrolyzing corn flour suspension with α-amylase alone for 3.0 h while supplementing with corn flour gave better results except a slight increase in DS and DB after the addition of β-amylase from malt extract or malt powder suspension (Table 3). To confirm the effect of β-amylase, reducing sugars obtained in the hydrolysate should be separated and analyzed. It was reported that the syrup produced by the maltogenic α-amylase would contain high level of maltotriose while the syrup produced by β-amylase would have large amount of β-limit dextrins and less maltotriose [2]. However in this studies it can be concluded that it is sufficient to use α-amylase alone to prepare a sugar syrup having DE between 50 and 70. To confirm this the sugar syrup was tested for its properties in industrial food preparations. When this syrup was tested for soft candy preparation the results were satisfactory. The syrup also should be tested in other food industries.

4 Conclusions

To obtain the sugar syrup of DE between 50 and 70, α-amylase alone is sufficient. Corn flour (22.0 %, w/w) suspended in tap water (pH 7.0) could be hydrolyzed first at 98 °C. At 1.0 and 2.0 h corn flour in suspension (5.0 g corn flour in 10.0 ml tap water) should be added and the hydrolysis should be continued for another hour.

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