



Minimizing of Dextran during Sugar Beet Manufacturing

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Authors' contributions

This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.

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ABSTRACT

The most serious processing problem can clear arise from the presence of dextran gum. The presence of dextran in sugar processing leads to less of sucrose and creates problems to sugar producers by increasing viscosity, lowing sugar yield, increasing molasses purities slowing filtration. The application of dextran enzyme to reduce dextran from raw juice was more efficient and economic than adding it to clear juice and syrup. Sixty percent of dextran removal was achieved when dextranase applied at concentration of 20u/100ml raw juice and 30min incubation. The dextran reduction and reached 65% by the use of 30u under some condition in clear juice, the percentage of dextran reduction reached 25,27 and 45% when dextranase enzyme used at 30u /100mol after 10,20 and 30 min of the incubation respectively. The use of the application of dextranase enzyme to reduce dextran from raw juice was more efficient and economic than adding it to clear juice and syrup.

Sixty percent of the dextran was removed when using dextranase applications at a concentration of 20u/100ml of raw juice over 30 minutes.

Keywords: Dextranase; raw juice; clear juice; harvest; syrup.

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1. INTRODUCTION

The utilization of dextranases in the sugar business was spearheaded in Australia during the 1970s. In South Africa where diffusers are utilized, dextranase application in diffuser stick juices was considered unacceptable for the most part due to the high temperatures [1]. As of late, new techniques have been accounted for to upgrade the enzymatic hydrolysis of dextran catalyzed by dextranase utilizing power ultrasonic joined with high hydrostatic pressing factor or microwave irradiation which could be a compelling strategies for improving the modern proficiency of dextranases in numerous mechanical applications including sugar fabricating measures [2, 3, 4]. The primary focal point of this exploration work was to examine the impacts of hydrolysis of dextran utilizing dextranase catalyst on the crystallization cycle and the nature of the last sugar gem. Besides, the impacts of expansion of dextranase to sugar squeezes as mechanically applicable strategy on development pace of sucrose gems, gem shape and surface geology at various crystallization temperature and supersaturation were likewise recognized [5] the starch and dextran present in cane juice meddle in their explanation. The degree of the presence of these poly saccharides in juices relies upon thickness, development period, collect and transport conditions. High substance juices show low filtration rate and helpless crystallization. The use of dextran protein has been upheld as of late starch is common constituent of stick squeeze however the presence of dextran in juices is credited to icing, bacterial pollution decay or harmed to cane [6]. They likewise study the use of microbial dextranase, detached from the filtrate of *Pectinomyces* culture, to eliminate dextran from stick juice. They tracked down that the movement of chemical dextranase was most extreme at 50 °c and pH 5.6 and was relative to fixation from 2.0mg to 10mg protein and it was straight more than 60 minutes. The current examination was done to consider the utilization of dextranase catalyst to decrease dextran from crude juice, clear squeeze and syrup [7]. The level of dextran decrease of syrup was not exactly of crude juice out clear squeeze at various degrees of protein one adding. It very well may be prescribed to add dextranase compound to push juice. Since the utilization of dextranase chemical to lessen dextran from crude juice was more productive and prudent then from clear squeeze and syrup [8].

2. MATERIALS AND METHODS

2.1 Materials

Dextranase enzyme from *paecilomyces lilaceous* (79.5u/mg) was obtained from fluke company, specialty chemical and analytical reagents for research.

2.2 Dextranase Treatments

Raw juice (brix 18.4, pH 5.5 and purity 86.4), clear juice (brix 18.03, pH 9.1 and purity 90.5) and syrup (brix 6.7, pH 9.1 and purity 90.5) were obtained for this study from delta factory. Dextranase enzyme was solved in acetate buffer solution at pH 5.5 and added at levels of 0,10,20 and 30u/100ml to 250ml of previous samples and mixed thoroughly for 40°c the samples /dextran mix was then immediately placed in water bath at 50°c. Aliquots (40ml) were removed after 0,10,20 and 30 min and were boiled immediately for 2.5 min. the samples were analyzed for dextran and viscosity after cooling.

2.2.1 Determination of dextran

Dextran was determined according to procedure of [8] as follows:

2.2.2 Reagents

- (i) Absolute ethyl alcohol
- (ii) ethyl alcohol 80%. dilute 80 ml of Absolute ethyl alcohol with 20 ml of distilled water [9]
- (iii) Sodium hydroxide solution 2.5n. dissolve 100g of Sodium hydroxide in water dilute to 100ml and saturated with Sodium sulfate.
- (iv) Copper reagent .prepare a stock solution by dissolving 3.6g of $\text{CuSO}_4 \cdot 4\text{H}_2\text{O}$ and 30.0g of sodium citric separately in small amounts of water, then mix and dilute to 1000ml with distilled water. to prepare the reagent dilute 50ml of the stock solution with 50ml of water of dissolve in this 12.5g of anhydrous sodium sulfate. The reagent must be freshly prepared every day.
- (v) Phenol solution 5% dissolve 5.0g of pure phenol in water of dilute to 100ml.
- (vi) Wash solution to 50ml of water add 10ml of copper sulfate reagent of 10ml of 205n Sodium hydroxide.
- (vii) Trichloroacetic acid solution 10% dissolve 10.0g of Trichloroacetic acid in water of dilute to 100ml.
- (viii) Concentrated sulfuric acid.
- (ix) sulfuric acid 2.0n dissolve 98g of Concentrated sulfuric acid in water of dilute to 1000ml.

Table 1. Standard curve

Stock solution ml	Dilute to ml	Dextran content mg/ml
10	100	0.01
20	100	0.02
30	100	0.03
40	100	0.04
50	100	0.05
60	100	0.06
70	100	0.07
80	100	0.08
90	100	0.09
100	100	0.010

2.3 Standard Curve

Weigh 500mg standard dextran in weighing dish and dry at 105°C for 4 hours. Cool in desiccators over anhydrous CaCl_2 weigh and calculate moisture.

Weigh another 500 mg dextran (corrected for moisture content), dissolve in water and dilute to 500ml. don't use predried dextran contains 1.0mg dextran 1ml.

Dilute 100ml of this solution to 1000ml. this solution contains 0.1mg dextran /100ml.

Aliquots of this dilution are diluted.

2.4 Phenol sulfuric acid test

In arrangement of 10 test cylinder 20×150mm spot 2ml of each dextran standard arrangement. Spot 2ml water in an eleventh cylinder as clear. Add 1ml 5% phenol answer for each cylinder. Twirl tubes tenderly to blend phenol and starch arrangement. Add 10ml concentrated sulfuric corrosive to each cylinder ideally utilizing plastic programmed pipet, is streetcar blended. Try not to allow pipet to contact arrangement, and don't allow corrosive to stay on top of arrangement. Blend on vortex blended.

Place tube (in rack) in boiling water bath for 2 minutes then let to cool for 30 minutes. Read color as absorbance (A) a transmission (T), prefer ably A, on color meter a spectrophotometer at 485 nm against blank solution set A to zero on blank and then read A of solution, using the seam cell (or matched cells) for blank of test solution. Make duplicate colorimetric readings of calculate average value. The color reading is then plotted on square paper against dextran concentration. The curve is used to determine the dextran corresponding to the color measurement in the unknown solution.

2.4.1 Preparation for dextran determination

2.4.1.1 Raw sugar

Precisely weigh 40g sugar in measuring glass and disintegrate in modest quantity of water. Quantitatively move answer for 100ml volumetric glimmer and weaken to volume with water, channel 50ml of this arrangement through subjective channel paper in pipe to eliminate coarse suspended material [10] Flautist 10ml filtrates in to 100ml outright liquor and mix. Let stand 5 minutes to from encourage (ppt) channel accelerate of on sintered glass channel on ring an elastic sleeve - topped vacuum flagon. Wash hasten multiple times with 80% ethyl liquor, filling channel with liquor each time and leaving liquor alone drawn through encourage. Try not to let accelerate to dry and liquor over stream channel. This progression is critical to eliminate sugar that may remain adsorbed to encourage and meddle in phenol – sulfuric response. When last portion of alcohol wash has been completely drawn through precipitate, quantitatively transfer precipitate plus filter acid to 25ml volumetric flask. Use minimum amount of water for transfer. First place 50 or 60 mm long -stem funnel in empty 25 ml flask. Invert sintered glass funnel containing precipitate on long -stem funnel full stem of sintered glass funnel with water of blow precipitate plus filter aid in to long stem funnel using air house. Use water from wash bottle to wash remaining traces of solid from sintered glass funnel (2 times) in to long -stem funnel of wash all precipitate (broken up with spatula) in to 25 ml volumetric flask. Amount water of used must not exceed volume of 25 ml. Adjust to with water Filter solution through fluted Whatman no 42(110 mm) paper in 60- or 80-mm funnel. Collected 10 ml filtrate for analysis. Pipet to 10 ml filtrate into 20×100 mm glass aplastic test tube. pipet 2 ml 2.5n NaOH reagent arrangement into test tube pipet 2 ml cu reagent arrangement in to test cylinder and add 0.2 g channel help.

Spot test tube (in position) in boiling water both 5 minutes to hasten cu dextran complex or channel helps and afterward let to cool for 20 minutes.

Filter precipitate which contain cu – dextran complex, on sintered glass filter (coarse poor size 15 ml). Rinse test tube 2 times with portions of wash solution and pour these into sintered glass filter. Discard flinty place filter containing precipitate, on vacuum flask counting short (325 ml) Nessler tube (or flat bottom Viet) positioned so that stem of sintered glass fanned extends into Nessler tube. Pour 2 ml 2 NH₂so₄ solution onto precipitate in sintered glass funnel turn on vacuum and draw acid solution through precipitate repeat this procedure then rinse precipitate with 2 ml water. Quantitatively transfer filters which contains solubilized dextran, to 25 ml volumetric flask and dilute to volume with water pipe 2 ml of this solution into 20×150mm test tube and follow procedure for phenol sulfuric acid test.

2.4.2 Beet juice

The brix of the juice is determined. Then 10 ml of juice is placed in 100 ml beakers 0.3-0.4g of analytical filter aid and 1 ml of 10% trichloro acetic acid are added to solution of mixed and 40 ml absolute are added. Some procedure as descended for sugar is then followed

2.4.3 Syrup

Weight 150g of syrup and dilute to 750g with water (1:5), then the brix is Determined and 10 ml is placed in 100 ml beakers and some procedure is followed.

2.4.4 Molasse

Weight 150g of molasse and dilute to 900 g with water (1:6), then the brix is Determined and 10 ml is placed in 100 ml beakers and some procedure is followed. Calculation of dextran as ppm/brix read dextran concentration mg 1ml from standard curve. Calculation dextran ppm (mg /kg) on slides as follows:

$$\text{Dextran ppm} = \frac{1}{A} \times \frac{1}{B} \times \frac{C}{D} \times E \times F \times 10^5$$

Where

A= weight of sample solids (g) diluted to 100 ml for or from sugar sample of brix for beet juice.

B= ml of aliquots taken for alcohol precipitin =10 ml

C= ml of solution of alcohol precipitate =25 ml

D = ml of aliquot taken for copper precipitate =10 ml

E = ml of final solution of copper -dextran complex = 25 ml

F = mg /ml dextran (from standard curve)

2.4.5 Viscosity

Viscosity of raw juice, clear juice and syrup were measured with a brook field DV-II rotational viscometer at 25°C. Where spindle no.18 was used the ppm rate applied was 150 according to [11].

3. RESULTS AND DISCUSSION

Table 2. Effect of dextran enzyme on viscosity of raw juice, clear juice and syrup after different incubation time. The effect of dextranase on viscosity reduction in the raw juice is illustrated in Table 2 at 0 u /100ml of raw juice and 0 /min of incubation time the viscosity was 1.79 cp. But after adding dextranase enzyme by 10u/100ml raw juice the viscosity was reduced by 8.37% ,15.08% and 20.11% after 10 ,20 and 30 min on incubation time respectively.

The percent of viscosity reduction of raw juice increased by applying 20 u 100ml raw juice at different time of incubation (10.05% ,40.22% and 50.27% at 10 ,20 and 30 min. reach to 32.40% ,46.36% and 52.51% when dextranase enzyme added by 30 u /100ml raw juice after 10 ,10 and 30 min incubation time respectively. From Table (2) it could notice that there are no significant different between control sample 0 u /100ml and 0 min incubation time and samples ferreted with 10 u /100ml at different concentration also, there are no significant difference between control sample and sample treated with 20 u / ml for 10 min. The highest viscosity reduction was achieved by the use of 30 u of dextranase enzyme for 30 min incubation time followed by 20 u/100ml for 30 min and 30 u/100ml of 20 min after different periods of incubation.

3.1 Effect of Dextranase Enzyme on Viscosity of Clear Juice

The effect of various levels of dextranase enzyme on viscosity of clear juice is illustrated in Table 2. no significant difference on viscosity reduction of clear juice was found when dextranase enzyme was used at 10 u/100ml for 10,20 and 30min.10u/100ml for 20min and 30u/100ml for 10min incubation time. Significant effect on viscosity reduction was found when enzyme applied at 20u/100ml clear juice for 20

and 30min as well as 30u/100ml for 20and 30min incubation time. Also, there are significant differences between treatment of 20u/100ml and 30u/100ml for 10,20and 30 min incubation time. Generally using 30u/100ml clear juice for 30min ,20u/100ml for 30min and 30u/100ml for 20min recorded the highest percentage of viscosity reduction (40.66%,38.83%, and 32.33% respectively).

3.2 Effect of Dextranase Enzyme on Viscosity of Syrup

From data in Table 2 it could see that using 10u/100ml syrup for 10min incubation time showed slightly viscosity reduction while 5.07% viscosity reduction was a sheared by extending the incubation time to 20min. Using 10u/100ml syrup for 30min incubation time improved the percentage of viscosity reduction to 10.03%. when the reaction occurs with 20u dextranase enzyme/100ml syrup 4.03% of viscosity was reduced off after 10min of incubation. Increasing of incubation time to 20min improve viscosity reduction was recorded by using 20u/100ml syrup for 30min.however, when dextranase

enzyme was applied at concentration of 30u/100ml the percentage of viscosity reduction reached 10.03%, and 30.10% after 10,20 and 30min of incubation. These results are in agreement with those of [11] who reported a significant reduction in viscosity was observed when 45pp/juice in final evaporator syrup of juice was used.

3.3 Effect of Dextranase on Raw Juice

The effect of addition various levels of dextranase enzyme of raw juice containing dextran (145ppm /brix) at 50 c is shown in Table (3).

It could be seen that 10 u/ml dextranase enzyme had very little on degradation with only 12% reduction after 10 minutes. Extending the incubation time to 20and 30 min.at the same concentration led to slight improvement of dextran reduction 25% and 30%, respectively. Treatment with 20u dextranase /100ml reduced 23%,50% and 60% of dextran after 10,20 and 30min respectively.

Table 2. Effect of dextranase enzyme on viscosity of raw juice, clear juice and syrup after different incubation periods

Enzyme Concentrate	Incubation time	Viscosity		
		Raw juice	Clear juice	syrup
0 u /100 ml	0 min	1.79	1.33	8.67
10 u /100 ml	10 min	1.64	1.33	8.49
	20 min	1.52	1.19	8.23
	30 min	1.43	1.06	7.80
20 u /100 ml	10 min	1.61	1.14	8.32
	20 min	10.7	0.97	7.36
	30 min	0.89	0.82	6.93
30 u /100 ml	10 min	1.21	1.06	7.82
	20 min	0.96	0.90	6.91
	30 min	0.85	0.79	6.06

Table 3. Effect of various levels of dextranase enzyme on dextran of raw juice after different incubation periods

Enzyme concentration (u/100 ml)	Incubation time (min)	Dextran concentration In raw juice ppm/Brix
0	0	145
	10	127
	20	108
10	30	101
	10	112
	20	70
20	30	70
	10	84
	20	63
30	30	51

Higher affect was achieved with 30u/100ml which removed dextran up to 42%, 56% and 65% after 10,20 and 30 min of the incubation respectively. These results are agreement with [11] Data in Table (3) demonstrated that these are highly significant between control samples and other treated samples. Whoever, no significant deference was found in level of dextran reduction of 10,20 and 30 min of incubation at deferent concentration of enzyme addition.

3.4 Effect of Dextranase on Clear Juice

As it can see in Table (3) only about 7% dextran had been removed at 10u/100ml dextranase enzyme after 10 mi. Incubation dextranase level to 20u/100ml slightly improved in dextran reduction by 14% after 10 min. incubation and even at 30u/100ml that only 25% dextran was removed from the clear juice at the same time of. incubation increasing of. incubation time to 10, and 20min.at 10u/100ml dextranase enzyme led to used sample difference of dextran removed (15% and 19% respectively) dextranase with 20u /100ml resulted in 31% removal of dextran after 20min and 38% after 30 minutes. While using of 30u/100ml dextranase for 20and 30min improved dextran reduction up to 27% and 45%, respectively. The effect of addition various levels of dextranase enzyme of raw juice containing dextran (145ppm/Brix) at 50°C is shown in Table (3).

3.4.1 Effect of various levels of dextranase enzyme on dextran of raw juice after different incubation periods

As it very well may be found in Table (4) just about 7% dextran had been eliminated at 10 u/100mil dextranase compound after 10 min. Expanding of dextranase level to 20 u/100mil marginally improved in dextran' decrease by 14%

after 10 min. hatching and even at 30u/100mil that solitary 25% dextran was taken out from the reasonable squeeze simultaneously of brooding, expanding of brooding chance to 10 and Tomin at 10 and 20min at 10u/100ml dextranase protein prompted utilized little distinction of dextran. Removal (15%and 19%; respectively) dextranase with 20u /100ml resulted in 31% removal of dextran after 20min and 38% after 30 minutes. While using of 30u/100ml dextranase for 20 and 30min improved dextran reduction up to 27% and 45%, respectively.

3.5 Effect of Dextranase Enzyme on Syrup

The effect of dextranase enzyme at different concentration on the syrup illustrated in Table (5).at 10u/100ml dextran removed was increased gradually across incubation time for 10,20 and 30min (2%, 5% and 8%), respectively. Higher dextran reduction was obtained at 30u/100ml of dextranase, where 10, 20 and 30min incubation resulted to dextran removed of 9%, 18% and 26%; respectively. These dextran reduction levels in syrup at 30u/100ml dextranase still less than those of dextran reduction in raw juice and clear juice at the same level of enzyme used. Table (5): show that there are no significant differences between central Sample (6u/100ml for 0 mon incubation time) all Samples treated with 10u/100ml for 10 min)0u/100mlfor 20 min and 20u/100ml for 10min. But there are significant differences between Central Sample as other treated samples. At different incubation time 10, 20 and 30 min the use of 10u/100 syrup show no significant difference. Table (5): showed that the percentage of dextran removal from raw juice is higher than from syrup at different concentration

Table 4. Effect of various levels of dextranase enzyme on dextran reduction of clear juice after different incubation periods

Enzyme concentration (u/100ml)	Incubation time (min)	Dextran concentration In clear juice ppm/Brix
0	0	166
	10	154
	20	141
10	30	134
	10	143
	20	114
20	30	103
	10	124
	20	105
30	30	91

Table 5. Effect of various levels of dextranase enzyme on dextran reduction of syrup after different incubation periods

Enzyme concentration (u/100ml)	Incubation time (min)	Dextran concentration In raw juice ppm/Brix
0	0	193g
	10	189
10	20	183
	30	177
	10	185
20	20	170
	30	153
	10	174
30	20	156
	30	144

of dextranase enzyme. This may be due to high concentration of solids in syrup. These findings are in agreement with results of [10] who found that applications of dextranase to juice were much efficient and economical than adding it to evaporator syrup. They announced additionally that in crude sugar factors. Solids are concentrated from around 15 Brix in juice under than 65 Brix to last evaporator syrup. As an outcome, the dextranase needs to follow up on in 4.5 occasions in view of much dextran in syrup than in juice. They expressed likewise that action of dextranase was steady up to 25-30 Brix, however after words diminished drastically. This is on the grounds that as the Brix increment there is a duplication of water action decline, which in the second substrate in the hydrolysis of dextran by dextranase protein. Another commitment factor might be the grouping of contaminations which restrain dextranase movement all however the brix of clear squeeze was not exactly the brix of crude squeeze, the level of dextran expulsion from clear squeeze was not exactly the level of dextran expulsion from crude juice at various fixations. This might be because of the great pH of clear juice (pH 9) contrasted and the pH of crude juice (pH 5.5). These outcomes were in concurrence with finding of [12] who detailed that ideal pH to finish the response was 5.5 PH.

4. CONCLUSION

Within the limits of this study, it was concluded that in sugar creation, dextran are bothersome mixtures blended by foreign substance microorganisms from sucrose, expanding the thickness of the stream and diminishing modern recuperation, achieving huge misfortunes. The utilization of the use of dextranase protein to lessen dextran from crude juice was more productive and monetary than adding it to clear squeeze and syrup. A little over half of dextran

expulsion as accomplished when dextranase applied at centralization of 20u/100ml crude juice and 30min hatching. The dextran decreases and arrived at 65% by the utilization of 30u under some condition in clear squeeze, the level of dextran decrease arrived at 25,27 and 45% when dextranase protein utilized at 30u/100mol after 10,20 and 30 min of the hatching separately.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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