



## Investigations of the influence of dextran on sugar cane quality and sugar cane processing in Kenana sugar factory

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### ABSTRACT

*In sugar production, dextrans are undesirable compounds synthesized by contaminant microorganisms from sucrose, increasing the viscosity of the flow and reducing industrial recovery, bringing about significant losses. In this article a laboratory investigation have been done on the concentration of dextran in kenana sugar factory deteriorated cane and sugar industry products, as well as their effects on the sugar factory operation. In addition, the effective concentration of biocides (Busan and formaldehyde) as inhibitor to bacteria responsible for formation of dextran has been done. Because of the spectrum of molecular weight of dextran the Robert and Polarization methods were used for juices and Haze method for quality of sugar. The results obtained by Robert method were the average concentration of dextran in crusher; mixed, clarified and limed juices are 603,358, 289, and 424 ppm respectively. Also the results obtained by polarization method show that the average concentration of dextran in crusher juice from filed (burned cane) was lower than dextran in crusher juice from factory. The results obtained by Haze for A- and B- raw sugar 56 and 86 ppm respectively. It was found that the high viscosity of dextran affected the rate of flocculation. It was noticed that pure juices had turbidity of 14.17; an addition of 1.6g dextran in 100ml of juice elevated the turbidity to 17.7nm, besides increase reading of Pol and Brix. It was found when 1 ml of Busan diluted 34 times with water while 1 ml of formaldehyde diluted 4 times, 0.05 ml from each was added to 1 ml of crusher juice both proved to be effective to inhibit the growth of bacteria. Accordingly comparing the biocides (Busan and Formaldehyde), it was found that the Busanis clearly more effective than Formaldehyde.*

**Key words:** Dextran, Sugar quality, Leuconostocmesenteroides, Sugarcane deterioration, Inhibitor

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### INTRODUCTION

Sugarcane industry is considered one of the organized sectors. This sector is among the countries leading economic enterprises. Sugar is mainly extracted from sugarcane and sugar beet. Studies have indicated that nearly 20-30% of total sucrose synthesized by sugarcane plant is lost during various stages of raw material handling and sugar mill processing. The post-harvest sugar loss is one of the most alarming problems of sugar industry and has attracted widespread attention in the recent years (Priyanka et al. 2010)

Polysaccharides are long chain molecules, either branched or straight. These molecules are derived from two sources :the metabolic activities of the growing plant (e.g .starch) and the metabolic activities of microorganisms (e.g .dextran) growing during its life or at some stage in the subsequent processing (James and Day, 2000).

Dextran is an extracellular bacterial homopolysaccharide of D-glucose composed predominantly of  $\alpha$ -1,6-glucopyranosidic linkages within the main chain. A polysaccharide usually referred to as dextran compound widely occurs in deteriorated sugar cane and beet. These molecules are derived from the metabolic activities of microorganisms growing during plant cultivation or at some stage in the subsequent processing (James and Day, 2000).

Dextrans can be responsible for problems in sugar processing which reduce both the recovery of sucrose during sugar production and the final quality of the sugar. Dextrans can be formed by many microorganisms and are not well-defined substances with specific properties. Dextran is the name given to a large class of extra-cellular bacterial polysaccharides composed almost exclusively of glucose units linked predominantly by 1:6 bonds, but also containing 1:4, 1:3 and some 1:2 glucosyl linkages. Dextrans in the sugar industry are predominantly linear, but (Edyeet al. 1995) have shown that branching can be significant, particularly with the low molecular weight dextrans where 5 to 8% branching was indicated. Dextrans in sugar processing occur as a result of post-harvest delay and, infrequently, as a result of poor factory hygiene. (Morel2002).

In sugar production, dextrans are undesirable compounds produced by contaminant microorganisms from sucrose (Jimenez, 2005). On contrast, dextrans are used in the manufacturing of blood plasma extenders, heparin substitutes for anticoagulant therapy, cosmetics, and other products (Alsop, 1983; Kim and Day, 2004; Leathers et al., 1995; Sutherland, 1996).

From an industrial point of view, studies have shown that *Leuconostocmesenteroides* strains are able to utilize high percentages of the sugar present in juices in a short time period. This implies important losses if the infection level is not controlled. High loss in product yield (sucrose) and contamination of the industrial process cause various problems such as: an increase of juice viscosity which produces blockage in the process line, pumps and filters; lower heat exchange; evaporation diminution; decrease in the efficiency and output of crystallization; crystal shape distortion; blockages in centrifuges and sucrose losses to molasses.

Dextran in juice, syrup and sugars can cause false polarization. Because dextran is highly dextrorotatory, approximately three times that of sucrose and gives a falsely high polarization, unless removed prior to test. Furthermore, high dextran levels reduce the efficiency of clarification techniques used in polarization determinations (Clarke et al., 1997; Cuddihy et al., 2000; Guglielmone et al., 2000).

In addition, the farmer is mainly paid based on the polarimeter reading, therefore, there is an obvious need for a dextran test in the core laboratory. The benefits arising from this would be the correction of the falsified reading and the identification of the sources of dextran contamination entering the factory (Singleton et al., 2001).

The problems associated with dextran contamination, in both the factory and the refinery, are well documented in the literature and thus are briefly summarized in Table 1.

**Table 1: Summary of the detrimental effects of dextran in terms of the losses it leads to according to Singleton et al., (2001)**

Production losses.	Sucrose losses	Direct financial losses
Increased viscosity leads to reduced throughput due to: ➤ Poor filtration rate ➤ Reduced evaporation rate ➤ Reduced flocculation rate ➤ Slow mud settling ➤ Poor crystallization (elongation)	Dextran formation  To molasses (melassigenic effect)	False polarimeter reading leads to overpayment to farmer  In trade of raw sugar as part of dextran penalty system using unreliable tests

This paper presents laboratory investigation on the concentration of dextran in kenana deteriorated cane and sugar industry products. The Robert and Polarization methods were used for juices and Haze method for quality of sugar furthermore comparative studies between three methods were carried out. In addition to the effective concentration of biocides (Busan and formaldehyde) as inhibitor to bacteria responsible for formation of dextran have been done. Also the effects of addition of purified cane dextran B-512 dextran on the turbidity, Pol and Brix of clarified juice were investigated.

## EXPERIMENTAL SECTION

### 2.1 Material

Samples :sugar cane slices, raw juice, clarified juice, thick juice and the final products, sugar were collected from Kenana sugar factory –Sudan

Dextrans: Three different molecular mass fractions of dextran (T40 :M~40,000 g/mol, product Nr .31389, T500 : M~500,000 g/mol, product Nr .31392 and T2000: M~2,000,000 g/mol, product Nr .95771)from Sigma-Aldrich were utilized .All fractions were produced by Leuconostocssp.

Chemical for analysis :all chemicals were obtained from Sigma-Aldrich, Germany.

## 2.2 Analytical methods for determination of dextran

In the sugar industry, several different methods are in use for the determination of dextran. These methods are Haze, Robert's copper and Polarimetric method.

### 2.2.1 Robert method

Dextran was determined according to Roberts, (1983) and AOAC, (1990). Roberts copper method determines dextran after polysaccharide precipitation from sugar solutions by 80 %ethanol. Quantification is made calorimetrically using Phenol- H<sub>2</sub>SO<sub>4</sub>, after a second precipitation with alkaline copper reagent .The amount of dextran  $m_{DE}$  in mg/kg DS in the samples was calculated as follows:

$$m_{DE} = \frac{1}{A} \cdot \frac{1}{B} \cdot \frac{C}{D} \cdot E \cdot F \cdot 10^5$$

- A Dry substance ( $w_{DS}$ )
- B mL of aliquots taken for alcohol precipitation (10 ml)
- C mL of solution of alcohol precipitates (25 ml)
- D mL of aliquot taken for copper precipitation (10ml)
- E mL of final solution of copper-dextran complex (25ml)
- F mg/mL dextran (from standard curve)

### 2.2.2 Haze method

The determination of dextran in sugar solutions by a modified alcohol Haze method is conducted according to the *ICUMSA* method (1994). The test sample is dissolved in water. Soluble starch is destroyed by incubation with a suitable enzyme (Novo Termamyl 120L, Novo Industri A/S, Bagsvaerd, Denmark). Protein is removed by precipitation with trichloroacetic acid (TCA) followed by filtration with acid-washed kieselguhr. The dextran haze is produced by diluting an aliquot of the treated, filtered solution to twice the aliquot volume by the addition of ethanol. The turbidity of the dextran haze is measured by reading the absorbance in a spectrophotometer at a wavelength of 720 nm. The method is standardized against a commercially available dextran.

This method measure the haze formed by dextran, like polysaccharides, when alcohols added to a solution of raw sugar.

The test sample was dissolved in water; soluble starch was destroyed by incubation with a suitable enzyme. Protein was removed by precipitation with trichloroacetic acid followed by filtration with acid –washed kiesselghur. The dextran haze was produced by diluting analiquot of the treated, filtered solution, to twice the aliquot volume by the addition of ethanol. The turbidity of dextran haze was measured by reading in spectrophotometer at a wave length of 720nm.The method was calibrated against a commercially available dextran.

The haze method is accepted commercially for dextran analysis. Therefore it was used in this study to determine the dextran concentration in raw sugar.

### 2.2.3Polarimetric method

Bose and Sinch (1981) reported a simple and quick method for estimation of dextran of the cane juice and they suggested the formula.

$$\Delta p = 0.001w - 0.1597$$

This is derived statistically by correlation dextran content, with the increase in the polarization value

$\Delta p$  = Polarization difference in pol reading.

W=dextran concentration (mg/kg).

### 2.3 Polarization

Polarization was determined by Automatic Saccharometer (Model: AUTOPOL 880, SN: 80851, USA) using a light source 589.44 nm (yellow) according to ICUMSA method GS1/2/3-1 (1994).

#### 2.4.1 Microbiological experiments

##### Isolation

All bacterial isolate used in this research were obtained from crusher juice, limes juice, clarified juice and syrup to obtain *Leuconostoc* S.P by preparing 8-fold serial dilutions of crusher juice in distilled water, and plating 0.05 ml on a modification of selective medium

Developed for *Leuconostoc* (Destefano 1986). The selective medium consisting of 23.5g Difco-Bacto plate count agar and 100 ml of raw sugar was dissolved in 1000ml of distilled water. Then the mixture was boiled on hot plate with a magnetic stirrer until it was dissolved, and then was autoclaved, and put in petridishes till it cooled.

The culture of bacteria was obtained by plating the above selective media with 0.05 ml of crusher juice, and then then the petridishes were put in an incubator for 48 hour.

The growth and non-growth was checked by 100xMicroscope after 48 hours.

#### 2.4.2 The addition of Busan to juices as inhibitor

Busan is a commercial name for L<sub>12</sub>-C<sub>14</sub> A/Kyldimethyl benzyl ammonium chloride (50% aqueous solution). It was used as inhibitor for *Leuconostocmesenteroides* in factories, 1 ml of Busan was taken and diluted to different concentrations, from each concentration 50ml was added to 1 ml of crusher juice in a test tube, and then mixed well. From the mixture 0.05 ml was taken and added to selective media as described in procedure above. Then after 48 hour the growth and non-growth was checked by 100xMicroscope

#### 2.4.3 The addition of formaldehyde to juices as inhibitor

The concentration of laboratory formaldehyde is 35% aqueous. The different concentrations of formaldehyde were prepared, and then the same procedure for the Busan was followed.

**2.5 Turbidity (IU)** -was determined according to SMRI Test methods TM025 (2004) as ICUMSA colour absorbance (720 nm) difference between unfiltered and filtered (0.45 µm cellulose nitrate membrane) solutions after dissolving sugar sample (50 g/100 mL) in distilled water.

#### 2.6 Brix

The mean °Brix of triplicate samples was measured using an index instruments TCR 15-30 temperature controlled refractometer accurate to ±0.01°Brix

## RESULTS AND DISCUSSION

As mentioned before there are many methods used for the dextran analysis. And because the spectrum of molecular weight of dextran ranges from a few thousand to millions, the Robert and Polarization methods are used for juices, and the Haze method for the quality of raw sugar. This is reflected in the results described below.

### 3.1 Roberts Method

This method is a quantitative method for dextran in which all polysaccharides are separated from the sugar and the dextran is selectively precipitated with alkaline copper sulphate. The dextran in the precipitate is then determined calorimetrically (Roberts, 1983) and (AOAC, 1990). Neither starch nor the indigenous sugar polysaccharides (ISP) is precipitated by copper sulphate and therefore does not react with the colorimetric reagents. The method is fairly rapid and the results are reproducible and independent of the molecular weight of the dextran. Because the dextran is separated from the sugar sample, this test is suitable for the use in dark coloured liquors and syrup.

Therefore this method was used for the determination of the dextran concentration in mixed juice, crusher juice, limes juice and clarified juice.

The method gave a good relation in calibrated curve between concentration and absorbance. The results obtained by Roberts method were shown in Table 2 and Figure 1. It was clear from the Table 1, the average concentration of dextran in mixed juice, crusher juice, limes juice and clarified juice are 358, 603, 424 and 289 respectively.

Also it should be noticed that, from Figure 1 the concentration of dextran in crusher juice cane was greater than others, this mean that the *Leuconostocmesenteroides* might have been active to produce dextran in milling, crush screen and cane carrier, and also in the previously damaged cane which is rich of dextran.

In addition, it was observed that the concentration of dextran in liming juice is greater than mixed juice and clarified juice. It is known that alkaline condition (pH-8) is the most favorable for the production of dextran at room temperature.

sugar contents in the final molasses increased in 0.6 points per 1 000 ppm of dextran in molasses, which is equivalent to 250 ppm in mixed juices, generating the loss of 0.6 pounds (0.272 kg) of sugar per ton of sugarcane(Efraín2005)

To this amount of sugar loss we must include the amount consumed for the formation of the 250 ppm of dextran: in the mixed juice, which according to prior data corresponded to 0.022 pounds (0.01 kg) per ton of sugar cane. That is, in the presence of 250 ppm of dextran in the mixed juice a total of 0.282 kg of sugar per ton of processed sugar cane was lost, which linearly extrapolated to the presence of 1 000 ppm of dextran in mixed juice generating a loss of 1.128 kg of sugar /ton cane.

Another study using conservative numbers from different studies around the world showed that each 0.1% increase of dextran in the juice (1000 ppm), resulted in the loss of 8.8 pounds (4 kg) of sugar per ton of sugar produced without considering the industrial recovery (Efraín2005). This implies the loss of an additional 0.77 pounds (0.35 kg) of sugar per ton of ground sugar cane assuming a recovery of 88%.

Another analysis for determining the losses of sugar caused by dextran showed a loss between 7.4 and 8 kg per ton of ground sugar cane(Efraín2005).

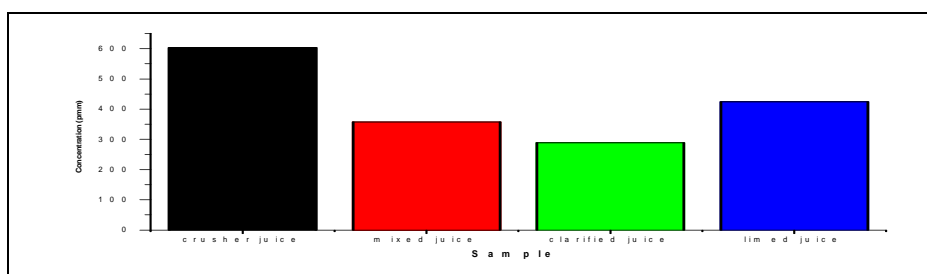
Other studies at more advanced stages of the process showed that for each 300 ppm of dextran in the syrup the purity of the molasses increased in 1% (Clarke et al 1997). Additionally it was shown that a 1 point rise in final molasses purity was equivalent to the loss of 0.454 kg of sugar per ton of sugar cane processed (Efraín2005)

The exact data on sugar losses caused by dextran are very difficult to determine since there are many influencing factors, starting from the distorted values of the initial sucrose contents, the variability between the methods used to determine dextran content and even the variation of the criteria assumed when analyzing this Rauh et al (1999).

In general, the results of all these studies show that the losses generated by dextran went from 0.35 kg of sugar per ton of ground sugar cane caused by the presence of each 1000 ppm of dextran in the mixed juice and reached 8 kg. Whatever the accuracy of this result, it shows the need to eliminate dextran from the sugar manufacturing process.

**Table 2: Average concentration of mixed juice, crusher juice, limed juice and clarified juice**

Sample	Brix	Absorbance	Concentration of dextran ppm
crusher juice	21.11	0.96	603
mixed juice	13.6	0.57	358
clarified juice	14.1	0.46	289
limed juice	14	0.68	424



**Figure 1: Relation between dextran concentration in mixed juice, crusher juice, limed juice and clarified juice**

### 3.2 Polarimetric method

The method is quick and simple, compared to haze and Roberts methods. Therefore it is used to check the deterioration of cane.

The results obtained by that method are shown in Table 3 and Table 4. It was noticed that the average concentration of dextran in crusher juice from field (burned cane) in Table (3) is lower than dextran in crusher juice from factory in Table (4) this means that the time delay between cutting and crushing increased the dextran concentration. This observation is in agreement with (Morel 2002) who mentioned that the dextrans in sugar processing occur as a result of post-harvest delay and, infrequently, as a result of poor factory hygiene

**Table 3: Dextran analysis by polarization method for crusher juice from field (burned cane)**

Pol reading by lead acetate	Pol reading by lead nitrate	Difference in pol reading	Dextran concentration ppm
38.89	38.57	0.3	379
35.43	35.25	0.18	339
34.20	34	0.2	359
30.05	39.7	0.35	509
34.86	34.76	0.1	259
34.37	34.23	0.14	299
41.70	40.54	0.16	319
38.08	37.94	0.14	299
31.98	31.88	0.1	259
39.12	38.92	0.2	359
36.42	36.28	0.14	299
37.36	37.32	0.04	199
<b>Average concentration of dextran (ppm)</b>			<b>301</b>

**Table 4: Dextran analysis by polarization method for crusher juice from factory (burned cane)**

Pol reading by lead acetate	Pol reading by lead nitrate	Difference in pol reading	Dextran concentration ppm
40.78	40.26	0.52	682
37.9	37.55	0.35	537
37.1	36.7	0.4	536
40.4	39.3	1.1	1209
31.08	30.8	0.26	422
39.7	38.5	1.2	1534
37.8	37.3	0.5	660
<b>Average concentration of dextran (ppm)</b>			<b>797ppm</b>

### 3.3 Haze method

The haze method is accepted commercially for the dextran analysis. Therefore it was used in this study to determine the dextran concentration. In A raw sugar the average concentration was 56ppm and 86 ppm in B sugar, as shown in Table 5. It was noticed that the concentration of dextran in A-raw sugar was less than concentration of B-sugar. Because the affination only removes 20%of dextran, the other 80%of dextran in the feed liquor is not adsorbed by carbonaceous adsorbent (Mead and Chen 1985)

Dextran is a polymer of glucose synthesized from sucrose by the action of microorganisms dextransucrase enzyme secreted by *Leuconostocmesenteroides*, *Streptococcus* and *Lactobacillus*(Kaur and Kaler, 2008; Aquino and Franco, 2009). The predominant microorganism implicated for dextran formation in the sugar cane industry is *Leuconostoc* species- ubiquitous in sugar cane field (Aquino and Franco, 2009). These enter the cane at places of exposed tissue caused by machine harvesting, cutting, burning, freezing, disease and pests. The presence of dextran indicates a lost sugar and is enhanced under wet conditions of temperature greater than 25<sup>0</sup>C. Dextran in the juice, syrups and sugars can lead to false pol reading (i.e., 1000mg/kg dextran can enhance pol reading by 0.30)(Kaur et al, 2008). Dextran concentrations greater than 500 mg/kg in raw sugar juice can cause processing problems, such as increased viscosity, slowed filtration, crystal distortion and sucrose losses (Kaur et al 2008) and Aquino, et al 2009). In the alcoholic and soft drinks industries, the presence of dextran could lead to the formation of haze and precipitations in the products and spoilage in other food industries, such as candy and chocolate manufacture (Aquino et al, 2009). In soft drink industry there is a suggestion to use dextran because of its use in drugs especially as blood plasma volume expander (Bhavani et al 2010).

Raw sugar with dextran concentrations above 250 ppm is subject to payment penalties of the magnitude of 0.007% of the price multiplied by the amount of tons sold. The fine value increases gradually in 0.002% with the increase in dextran concentration every 160 ppm, until reaching 0.013% for the contents equal or higher than 1 010 ppm (Efrain2005).

Table 5: Dextran analysis by Haze method for A- raw sugar and B- raw sugar

A-Raw Sugar		B-Raw Sugar	
Absorbance	Dextran concentration ppm	Absorbance	Dextran concentration ppm
2	12	2	18
3	18	5	32
1	6	19	116
6	37	9.3	56
5	31	9	58
6	37	7.7	47
13	50	21	131
16	100	14.7	92
15	94	4.5	28
9	58	17	107
10	66	6	38
4	25	25	157
12	75	25	157
6	38	28	176
17	110	-	-
9	53	-	-
23	145	-	-
<b>Average conc. of dextran</b>	<b>56ppm</b>	<b>Average conc. of dextran</b>	<b>86ppm</b>

### 3.4 Comparison between Haze, Robert and polarization methods

This section compares the three methods and explains the reasons why each one was used. The wide range of dextran molecular weights leads to the use of these three methods. When the Robert method was used, the molecular weight of the dextran did not affect the curve. This is because the phenol-sulphuric acid does not react with dextran, but the hot acid hydrolysed the dextran to glucose which reacted with the reagent to produce the colour. For this reason, this test determines all glucose units in the dextran and therefore the analysis was independent of the molecular weight of dextran used as standard.

Roberts(1983) showed the recovery by the copper and haze methods of dextran T2000 added to solution of refined sugar containing no measurable dextran. The recovery by the copper method ranged from 99% to 102% while the recovery by the haze methods ranged from 60 to 90%. In addition he also showed the recovery by two methods of dextran T40 (low molecular weight), the recovery by the copper methods ranged from 96% to 103% while the recovery by haze ranged from 49-73%.

The above observation indicates that the haze method gives lower results for dextrans of lower molecular weight. Also in Robert method, starch and indigenous sugar cane polysaccharides do not interfere in the determination. But in haze method, starch interferes in the determination. Therefore  $\alpha$ -amylase was used to remove starch.

However, unfortunately the current methods to determine dextran at the factory all have drawbacks. The two methods are tedious, costly and time-consuming and the Robert method consumes more reagents than the haze method.

The two methods need at least 2hours to determine the concentration of dextran in one sample. But now the haze method is used as a commercial method for raw sugar and not for juice.

If the two methods above are compared with the polarization method, there is a big variation between them. The polarization method is based on difference in pol reading between one sample clarified with lead acetate and basic lead nitrate. The method is simple and does not consume much reagent. Some study mentioned that it was used for testing badly deteriorated cane.

Generally the method is simple and needs less time than the two other methods

### 3.5 Turbidity

Strong correlation was observed between turbidity and color since as color increases, the non-sugar contents that contributes to the turbidity also increases. Turbidity is one main parameter used to assess the clarification process performance because it is related to the presence of non-sugar, flocks and suspended formation contributing materials such as starch, dextran and other indigenous sugar cane polysaccharides, gums and proteins in the juice (Hamerski et al. 2012). The removal of turbidity indicates the removal of these components. Soft drink bottlers demand non-foaming sugar without turbidity and the plantation white sugar from the three sugar factories are limited in this aspect to meet the requirements of soft drink industries.

The viscosity of the solution during clarification reduces the precipitation speed of impurities, forming scale deposits and decreasing the heating efficiency of the flow, thereby generally impoverishing the process. The corresponding juice derived from the deteriorated sugar cane with more acid pH values, consumed larger amounts of lime for its neutralization, hence providing higher turbidity, generating a larger volume of sticky mud that causes press filter blockage (Imrie et al 1972).

Similar results were found when different concentrations of dextran were added. That is clear in Table 5 and Figure 2. From the Table 5 it is noticed that pure juices have turbidity of 14.17, and addition of 1.6 g dextran in 100ml of clear juice elevates the turbidity to 17.7. This affects the rate of flocculation, floatation rate of coagulum and rate of scum compression leading to a low sugar quality.

**Table 5: effect of dextran on Turbidity, Pol, and Brix**

Dextran added to clarified juice	Brix	Pol	Turbidity at 900nm
Pure juice	12.8	48.5	14.17
0.2g dextran per 100 ml clarified juice	13	49.7	14.9
0.4g dextran per 100 ml clarified juice	13.3	51.1	15.3
0.6g dextran per 100 ml clarified juice	13.5	52	15.8
0.8g dextran per 100 ml clarified juice	13.8	52.6	16.37
1g dextran per 100 ml clarified juice	14.1	53.3	16.6
1.2g dextran per 100 ml clarified juice	14.2	54.2	16.7
1.4g dextran per 100 ml clarified juice	14.5	54.4	16.9
1.6g dextran per 100 ml clarified juice	15.9	54.7	17.7



**Figure 2: Relation between dextran concentration and Turbidity**

### 3.6 Polarization

Dextran is dextrorotary having a specific rotation of at least three times that of sucrose. From Table 5 and Figure 3, it is noticed that the addition of dextran to pure juices increase the pol reading. This is because the dextran was dextrorotary and its specific rotation is almost three times that of sucrose. Clark (1983) showed that pol elevation created by dextran in cane juice (200ppm) was 0.6% (by pol). The sucrose loss due to dextran formation (2000ppm) was 0.4% sucrose.

The economic losses caused by dextrans are continuous throughout the process, since its early content in the juices falsely increases the amount of sugar calculated for them and alters the production indicators of the factory. This is due to the dextrorotary characteristic of dextrans that polarize approximately three times more than sucrose producing a high false Pol value (Cuddihy et al [www.midlandresearchlabsinc.com/doclib/ polysach.pdf](http://www.midlandresearchlabsinc.com/doclib/polysach.pdf)).

A study performed by adding standard dextrans to pure sucrose solutions showed that for each 180 ppm of the polysaccharide there was a mean increase of polarization of 0.05 °S (Efraín et al 2005).



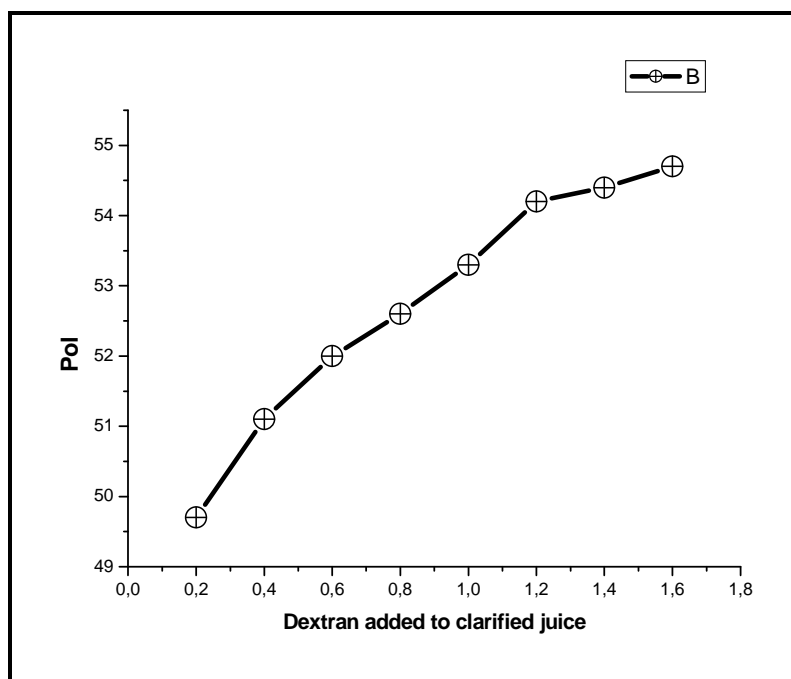


Figure 3: Relation between dextran concentration and Pol

### 3.7 Effect of dextran on Brix

Brix is defined as total solid in juices. Therefore, the addition of standard dextran to clear juice increases the total amount of soluble solid as shown in Figure 4 and Table 5.

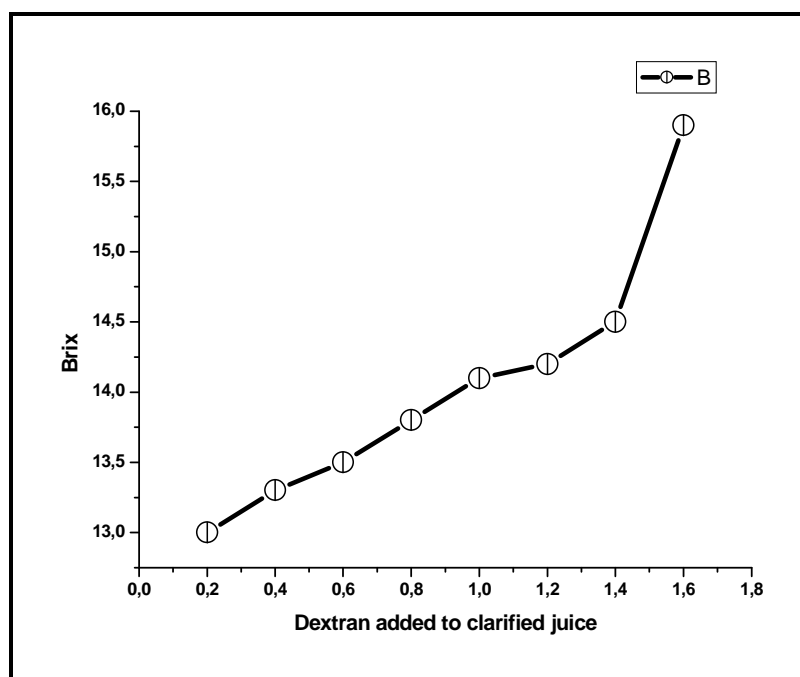


Figure 4: Relation between dextran concentration and Brix

### 3.8 *Leuconostoc Mesenteroides*

The inversion of sucrose by enzyme continues till it is heated during clarification. The prevention of sucrose inversion loss by enzymes and the control of the microorganisms responsible for continuous secretion of the enzymes in juice system is possible by good housekeeping and treatment of juice with an efficient biocide and effective concentration.

Good housekeeping alone does not prevent microbiological activities in the circulating juice, but also requires the treatment with an efficient biocide which disinfects the surfaces in the juice handling system, that are inaccessible to stream of hot water washing.

Among the disinfectants, the most widely used are C. M. A Busan, Chlorine, formaldehyde Upadhiaya, (1987). And hypochlorite (Bose *et al* 1970), In addition, various other substances like benzoic acid, hydrazides, sodium silicate (Ravelo *et al* 1991), Isothiocyanate, Polycides (dichlorophene in NaOH solution), and DNDT (Ravelo *et al* 1991) have also been tried. Moreover, even the use of gamma radiation as a cane sterilizer (Acosta *et al* 1982) has been recommended.

But to use any disinfectant for this purpose, its cost must be little, it must be highly efficient for use at low concentration and finally it must not pollute the environment.

*Leuconostocmesenteroides* the dominant microorganism in standing burnt cane after chopped-harvesting and transporting to the mill (Foster 1980).

Fortunately *Leuconostocmesenteroides* does not grow at temperature above 50°C or at high Brix (Morotz) and it grows favourably at PH 4.8 -6.0, and sucrose concentration 8-15%. The *Leuconostoc* is active between burning, cutting and crushing and particularly in chopped harvested cane, and at alkaline condition (PH-8) its most favourable condition for the production of dextran is at room temperature.

The sample taken from crusher and limed juice was found favorable to growth of bacteria. But there was no growth of bacteria in clarified juice and syrup, because the high temperature in the evaporator and pan boiling destroyed the bacteria.

### 3.9 Addition of Busan and Formaldehyde as Inhibitors

Once dextrans are present in the sugar production process the viscosity of the solution increases depending on the concentration and the molecular weight or the polymers formed which may range from 10<sup>5</sup> to 10<sup>7</sup> or more. The dextrans of a very high molecular weight are insoluble, while those of low molecular weight and soluble, are the ones that cause more difficulties in the production process.

The control of dextrans in the sugar industry is carried out by the rigorous adjustment between the burning, when performed, the cutting, mechanical or manual, and the delivery of the fresh sugar cane to the mill. Sanitation techniques of the technological equipment with steam every 8 hours during the sugar mill operation and the use of biocides on the sugar-cane in tandem are also executed. Any delay of over 14 hours in the arrival of the cut sugar cane to the mill under warm and humid conditions, favors the formation of dextrans, which will reach the mills and enter with the juice into the industrial flow. Dextran content increases progressively along the process from the dilute juice to the final molasses. The harmful effect of dextrans begins at their formation due to the irreversible sucrose consumption they produce. A study to evaluate these sucrose losses showed that the presence of 0.05% dextrans in raw sugar consumed 0.2 kg/t of sugar or 0.02 kg/t of processed sugar cane (Efraín (2005)

Recent studies showed that a *L. Mesenteroides* strain isolated in a sugar mill in Argentina during the first 6 hours of culture at 30 °C consumed sucrose at a rate of 8.46 g/L/h. The sucrose consumption reduced with an increase in temperature.

A significant loss of sucrose may occur when the bacteria *Leuconostocmesenteroides* become established in the milling process, this particular species of bacteria is capable of very rapid growth under the favorable conditions which usually exist in cane juice prior to heating (Upadhiaya 1987).

Therefore, to prevent microbial activities in circulating juice, it must be treated with an efficient biocides. The commercial biocides used are Busan and formaldehyde.

It was found that Busan was more effective than formaldehyde 1ml of Buasn was diluted 34 times and 0.05 ml of dilute Busan was added to 1ml of crusher juice. It proved to be effective to inhibit the growth of bacteria *Leuconostocmesenteroides* shown in Figure 5.

On the other hand 1 ml of formaldehyde was diluted to 4 times, and 0.05ml of the diluted formaldehyde was added to 1 ml of crusher juice, it was found sufficient to inhibit the bacterial growth.



**Figure 5: Micrographs of strains of *Leuconostoc* spp. by scanning electron microscopy**

## CONCLUSION

The loss of sucrose at all stages from sugar cane in the field crystal sugar in the bag, is a serious economic problem to the sugar industry, increasing costs and decreasing availability of land make it necessary for the industry to cut the losses to the lowest possible level.

The microbiological deterioration of sugar juices can lead to the formation of high –molecular weight polymers of glucose known as dextran.

Polysaccharides, like dextran, do not only reduce the quality of sugar, but cause difficulties in the sugar factory from milling to crystallization.

The objectives of this research was to investigate the influence of dextran in deteriorated cane and the quality of sugar produced thereof and the effect of dextran on sugar factory operation as a whole. So that efficient procedures can be applied to reduce the content of dextran in sugar cane produced.

Different methods were used to determine the concentration of dextran in sugar cane and other factory products. Two of these methods are Roberts and polarization methods. These methods were used to determine the concentration of dextran in crusher juice (from field), crusher juice from factory, mixed juice, lime juice and clarified juice. It was found that the concentration of dextran was highest in crushed juice from factory followed by limed juice, mixed juice and clarified juice. The results obtained by Roberts Method as average concentration of dextran in crusher juice, mixed juice, clarified and limed juices of Kenena cane farm were 603,358,289 and 424 ppm respectively. This means that the delay between cutting and crushing increases dextran concentration.

Also the results obtained by polarization method for the average concentration of dextran in crusher juice from field (burned cane) for the same cane were lower than dextran in crusher juice from factory.

In addition, Haze method was used for the determination of the dextran concentration in A-raw sugar and B- raw sugar. It was found that the average concentration of dextran in A raw sugar was 56ppm while in B-sugar was 86 ppm. It is clear that the average concentration of B-sugar was greater than A-raw sugar, which indicates that the dextran is not removed by affination process, but it passes through to the final product.

It was found that the high viscosity of dextran affected the rate of flocculation, it was noticed that pure juices had turbidity of 14.17 nm, and addition of 1.6gm dextran in 100ml of juice elevated the turbidity to 17.7 nm. Also addition of dextran to pure juice increased pol reading and gave false reading in polarization with increase of the Brix.

All these dramatically affect solution properties and sugar processing. They lead to quantitative and qualitative losses in sugar.

In order to decrease the formation of dextran by inhibiting the growth of microbes responsible for formation of dextran some trials were performed using formaldehyde and Busan.

It was found when 1 ml of Busan diluted 34 times while 1 ml of formaldehyde diluted to 4 times and 0.05 ml from each were added separately to 1 ml of crusher juice both proved to be effective to inhibit the growth of bacteria. It can be concluded that the Busan is more effective than formaldehyde.

Therefore, in order to reduce harmful dextran effects, it is necessary either to use effective biocide to inhibit the bacteria (*Leuconostoc*) as a main source for dextran, or use enzyme (dextranase) to hydrolyse dextran to lower molecular weight.

The failure of kenana practice to inhibit the growth of bacteria by adding Busan can be attributed to inefficient procedure of random addition of Busan. It can be shown by simple calculation that if 1ml of Busan is added to 680 ml of juice will be more effective in inhibiting growth of bacteria.

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