

Removal of Acid Beverage Floccs in  
Crystal Sugar by Adsorption Column  
Chromatography: Preliminary Study  
with Adsorbent Resin

Roberta Bergamin Lima and Claudio Lima de Aguiar\*

*Hugot Sugar Technology Laboratory, Luiz de Queiroz College of Agriculture, University of São Paulo, Brazil*

## Article Information

Received date: April 16, 2015

Accepted date: May 25, 2015

Published date: June 12, 2015

## \*Corresponding author

Claudio Lima de Aguiar, Hugot  
Laboratory Technology Sucroderivados,  
University of Sao Paulo, Brazil, Tel: +55-  
19-3429-4132; Fax: +55-19-3429-4275;  
Email: claguiar@usp.br

Distributed under Creative Commons  
CC-BY 4.0

**Keywords** Sugar; Adsorption;  
Chromatography; Color; Acid beverage  
flocs

## Abstract

The sugar industry has been facing some problems with white sugar intended for soft drinks production. Some studies have been demonstrated the sugar could be source of insoluble substances technically called by Acid Beverage Floccs (ABF). It appears during the beverage production; however, the floccs are easily disappearing with shaking. This may be related to raw material the sugarcane quality and, consequently, with the efficiency of the juice clarification process, since if there is no adequate juice treatment. The sugarcane commonly has lower quality and higher intensity of impurities, such as proteins, wax, minerals and colloids. The sugar industries are implementing solutions to reduce the amount of impurities through using technologies like ionic resins to removal of color, odor or undesirable taste. It could transform raw sugar in refined or white sugars. The aim of this work was evaluate the efficiency of purification of sugar solution with chromatographic adsorption system. We observed that the color reduction was around 69% and an intensive reduction in turbidity of sugar aqueous solution. There were variation on sucrose and increase on reducing sugars (glucose and fructose) contents. It is possible to conclude that the process could be efficient to reduce the color (MAU color), turbidity and ABF into sugar solutions.

## Introduction

According to Rosa [1], Brazil is the third largest exporter of soft drinks in the World after the United States and Mexico. Soft drinks, chocolate and ice cream manufacturers are responsible for approximately 55% of Brazilian sugar consumption [2]. The sugar industries are implementing solutions to reduce the amount of impurities from sugarcane and raw sugar through the use of technologies such as ionic resins for the removal undesirable color, odor and taste.

For Clarke et al. [3], Acid Beverage Floccs (ABF) has beginning with the formation of an intermediate floccs, through the possible interaction of proteins with polysaccharides (ISP). Further, there is interaction with the starch, dextran, silicate, colloidal substances and other soluble polysaccharides. The glucuronic acid from ISP and the primary amine residues are oppositely charged at the pH of beverages and charge through attraction, combine to form a coacervate as the basis for forming a flocc network. Other compounds may influence the formation of these insoluble substances.

In beet sugar, the saponins are responsible for the flocculation in acid drinks. For sugarcane, some efforts have been done to identify compounds related to the formation of ABF lead to analysis of various impurities such as starch, wax, protein, silicon dioxide and ashes. It has been found that acidic amino acids in carbonated beverage floccs are directly connected with the potential to flocculate sugar. They concluded that amylase together with other components, in particular proteins, were potentially responsible for the flocc appearing [4].

Given the importance of the sugar market and analytical industrial control, the aim of this work was to evaluate the effect of ABF removal process by adsorption chromatography system, evaluating the effect on MAU color, turbidity, sucrose and reducing sugars on sugar solution and the removal capacity of compounds that can influence the production of acid beverage floccs.

## Material and Methods

## Materials

The samples used were white sugar obtained from mills at Piracicaba region, Brazil. All the analyses were performed in Hugot Sugar Technology Laboratory, Luiz de Queiroz College of Agriculture, University of São Paulo, Piracicaba, SP - Brazil. All the reagents (3,5-dinitrosalicylic acid and acetonitrile) and known standards (sucrose, glucose and fructose) were purchased by

**Table 1:** Product specifications of adsorbent resin used by syrup treatment.

Parameters	Descriptions*
Matrix	Macroporous styrene-divinylbenzene copolymer
Functionalgroup	Tertiaryamine
Typicalsurfacearea	800 m <sup>2</sup> /g L

\*Dow (2015)

Sigma-Aldrich Co. (São Paulo, Brazil). The water used to prepare the sugar solutions at 65% (w/w; named syrup) was Milli-Q grade (Millipore Co.; São Paulo, Brazil). The resins specification used in this study were showed on Table1.

### Experimental procedure

**Column preparation:** The chromatographic assays were carried out in a vertical glass jacket column (dimension: 1” φ × 35.43” L; FGG Ltd., Brazil) and working volume of 455.2 mL. The 70% of the internal volume was filled with syrup and 30% of adsorbent resin (Table 1), about 136.06 mL (142.21 g resin with density equal to 1.04 g/mL). The assay was carried out at 65 to 70°C by recirculating water into a jacket column.

**Sample preparation:** The syrup used was 65 to 70° Brix (% of soluble solids (w/w)). Each volume pass through the column bed was sampling (50 mL) for further analyses.

**Column regeneration:** The resin was washed with Milli-Q water and NaCl at 10% (w/v). The resin was dried in an oven at 65°C.

### Sample characterization

**Absorbance million unit color:** The spectrophotometric color was measured by the ability of light passes through in a sugar solution at pH 7.0 and 420 nm. The values were expressed by Million Absorbance Million Unit (AMU) or the wavelength at which it absorbs the most light, and the concentration in parts-per-million.

**Reducing sugar analysis:** The reducing sugar contents were measured by the 3,5-dinitrosalicylic acid (3,5-DNS) method by Miller’s method [5]. It was used 1.0 mL of the diluted sample to 1.0 mL of DNS solution. The mixtures were heated up for 5 min in a boiling water bath and then cooled under running tap to adjust at room temperature. The color intensities were measured at 540 nm in UV Mini-1240 spectrophotometer (Shimadzu Co., Japan).

**Turbidity analysis:** Turbidity (expressed by NTU – Nephelometric Turbidity Unit) was evaluated in digital turbidity TB-10000Model (Tecnopom Co.; Piracicaba, Brazil). To fill the bucket turbidimeter was used Pasteur pipette with 3 mL in an attempt not to withdraw the sample particulate matter [6].

**Chromatography analysis of sucrose:** The method employing an UFLC chromatographic system (Shimadzu Co.; Kyoto, Japan) equipped with ELS-LT (Evaporative Light Scattering at Low Temperature) detector was carried out at 35°C using isocratic elution of acetonitrile (HPLC grade; Tedia Co., USA) and deionized water (Millipore Co., Brazil) at a flow rate of 1.0 mL/min and gain at 3.0. Isocratic elution was employed for 12 min with a mixture of 70:30 (v/v) acetonitrile-water. Nitrogen (≥99.0%; Air Liquide Co., Brazil) at 350 kPa was used to nebulize the effluent coming from the

**Table 2:** Sucrose, reducing sugars and °Brix for samples after adsorption treatment.

Samples	Sucrose(mg mL <sup>-1</sup> )	Reducing sugars(mg mL <sup>-1</sup> )	°Brix
Control	721.4±59.8 <sup>c</sup>	0.47 ±0.03 <sup>a</sup>	65.1 ±0.06 <sup>a</sup>
1° bedflow	853.4 ±5.1 <sup>a</sup>	1.97 ±0.09 <sup>b</sup>	63.7 ±0.12 <sup>a</sup>
2° bedflow	814.7±9.1 <sup>a</sup>	1.82 ±0.07 <sup>b</sup>	65.5 ±0.12 <sup>a</sup>
3° bedflow	739.4±7.5 <sup>bc</sup>	1.77 ±0.06 <sup>b</sup>	64.2 ±0.06 <sup>a</sup>
4° bedflow	806.87±2.5 <sup>ac</sup>	2.01 ±0.10 <sup>b</sup>	64.9 ±2.25 <sup>a</sup>

Results expressed by n = 3 ± standard deviation, and means followed by the same in the column do not differ statistically by Tukey test at 5% significance level.

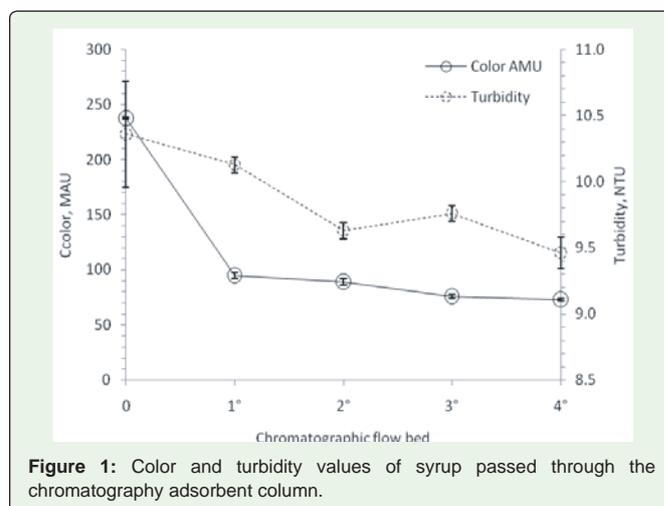
column NH<sub>2</sub>P-50 4E (250×4.6mm) Shodex Packed (Japan) and the evaporation temperature of the chromatographic eluent was 30°C. It was injected 5 µL of the sample diluted 0.5 mL in a volumetric flask at 25 mL [7].

**Brix analysis:** Brix degree of the samples were measured with Tecnal AR-200 digital refractometer at 20°C and expressed in °Brix (% soluble solids, w/w) [8]. (Spencer and Mead, 1945). For those assays was stored for a time and °Brix of syrup was accompanied by until it obtained the initial °Brix. Therefore, the soluble solids (°Brix) were used as a parameter control. Chromatographic column was conditioned with syrup solution until the sample taken with the same °Brix and thereafter the test started to be driven.

### Results and Discussion

Samples were submitted for four bed flows and samples was collected for color, reducing sugars, sucrose, °Brix and turbidity analyses (Table 2). According to Gokmenand Serpen [9], liquid phase adsorption processes have been shown to be highly efficient for the removal of color, odor, organic and inorganic pollutants.

There was a statistically significant reduction of syrup color against the control sample (without treatment by the adsorbent resin). Thus, the samples subjected to the third and fourth bed flows were not statistically different at p<0.05 (Figure 1). Diphenyl-styrene copolymer resins could form a strong link with the aromatic hydrophobic bodies of color compounds, by Vander Waals forces, creating a higher ability for adsorption as compared with other kinds



**Figure 1:** Color and turbidity values of syrup passed through the chromatography adsorbent column.

of resins [10]. We observed color reduction in samples submitted to chromatographic column compared to control samples.

Color Reduction Rates ( $CRR = ((\text{final color} - \text{initial color})/\text{initial color}) * 100$ ) were also calculated. Moreover, it was observed that the highest CRR (-69%) was found in the sample submitted for fourth bed flow. Each bed flows present color reduction in the syrup. There was also a significant reduction on turbidity values. The turbidity measures the light-scattering properties of a solution, consequently providing the most direct measure of the concentration of particles in a solution [11,12] that could be related with the reduction the potential floc formation. Besides that chromatographic method of removing colored compounds also was observed the reduction of turbidity samples. The sample turns more transparent and translucent.

Gokmenand Serpen [13] reported that an improvement of 40 to 60% in apple juice color undergoing adsorbent treatment for 2 h at 40 to 60°C. Serpen [9] reported that the adsorption was efficient to removal of Maillard reaction pigments, showing its efficacy in removing colored compounds from sugar solutions. Once the Maillard reaction products could developed a sequence of chemical reactions to form colored compounds, which disqualify the sugar because it turns dark. Maillard's reactions do not only form melanoidins, as well as, several volatile and flavor (aldehydes, ketones and pyrazine) via intermediates, such as 5-Hydroxy Methyl Furfural (5-HMF), reductones, aldimines and others [14].

Concern with the formation of reducing sugars was related to losses in sucrose. There was a gradual increase of reducing sugars (Table1), between the control and that samples passes through resin treatment. There was significant difference (at 5% probability) between the samples passed successive times on the chromatographic column. As for sucrose contents there were non-regular behavior, since the control sample showed lower sucrose content compared with other samples. Sucrose was been retained in the column and it been released in each bed flow. There were significant differences in control sample against the first and second bed flow samples and, those samples in relation with the syrup samples submitted for the third fluidized bed. The system was not homogeneous, demonstrating that there was not equilibrium as described by Kuan et al. and Azizian et al. [15,16].

Figure 2 presents the presence of acid flocs was observed after regeneration of column by washing with NaCl at 20% (w/v). According to Dow [17], although the treatment was efficient, the regeneration of the resin with NaCl that was not the more adequately process, that should be done with NaOH 4 % became de process more efficiently.

## Conclusion

Chromatographic adsorbent treatment showed efficient for acid beverage flocs from sugar solutions and it reduces significantly the color and turbidity values. The treatment did not showed excessive losses in sucrose levels, although there is an increase in reducing sugar contents. After the treatment, the presence of floc was observed during the washing and regeneration of resin.

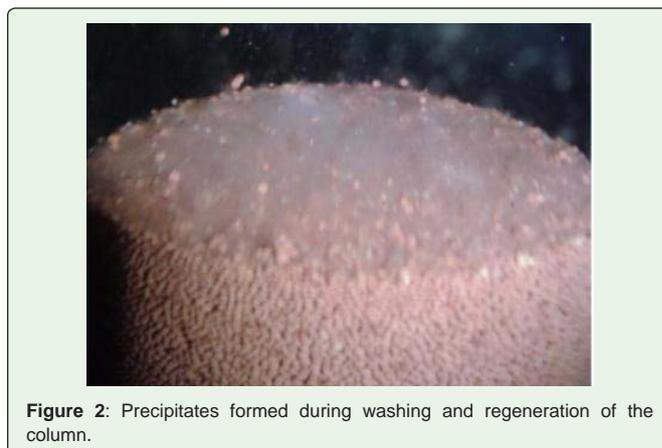


Figure 2: Precipitates formed during washing and regeneration of the column.

## Acknowledgement

The authors were thankful by financial supports from FAPESP #2009/54635-1, CNPq #310367/2013-1 and, finally to CAPES by provide funding to R.B. Lima. The authors were also thankful by technical support from Dow Chemical team.

## References

- Clarke MA, Legendre RB. Qualidade da cana-de-açúcar: Impactos no rendimento do açúcar e fatores da qualidade. STAB. 1999; 17: 36-40.
- Copersucar. Centro de tecnologia de cana. Manual de controle químico da fabricação de açúcar. Piracicaba. 2001; 261.
- Dow water and process solutions. 2015.
- Gokmen V, Serpen A. Equilibrium and kinetic studies on the adsorption of dark colored compounds from apple juice using adsorbent resin. J Food Eng. 2002; 53: 221-227.
- Hodge JE. Chemistry of browning reactions in model systems. J Agric Food Chem. 1953; 1: 928-943.
- Infosucro. Sistema agroindustrial, inovação e competitividade instituto de economia.
- Konen JC, Wilson JR. Replacing carbonaceous adsorbents with acrylic and styrenic Strong base anion resins in cane sugar decolorization applications. Intl Soc Sugar Cane Technol. 1992; 5: 1-7.
- Landi S, Mantovani G. Ion exchange in the beet-sugar industry. Sugar Technol Reviews. 1975; 3: 1-67.
- Liuzzo JA, Wong CM. Detection of floc producing sugars by a protein dye-binding method. J Agric Food Chem. 1982; 30: 340-344.
- Miller GL. Use of dinitrosalicylic acid reagent for determination of reducing sugars. Anal Chem. 1959; 31: 426-428.
- Rosa SES, Cosenza JP, Teixeira LS. Panorama do setor de bebidas no Brasil. 2014.
- Serpen A, Ataç B, Gokmen V. Adsorption of Maillard reaction products from aqueous solutions and sugar syrups using adsorbent resin. J Food Eng. 2007; 82: 342-350.
- Spencer GL, Meade GP. Special reagents. In: Cande Sugar Handbook. Wiley: New York. 1945.