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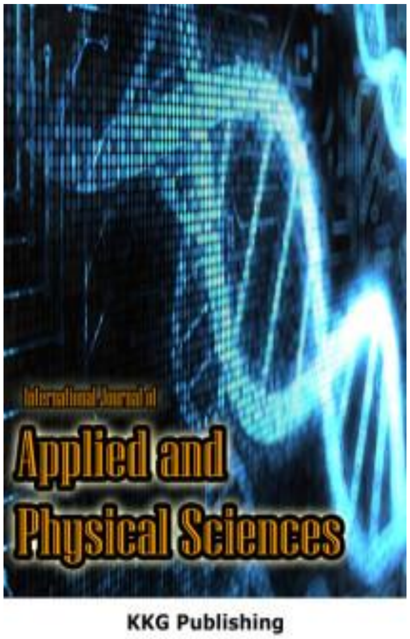

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EXTRACTION OF RESIDUAL SUGARS FROM SWEET PEARL MILLET AND SWEET SORGHUM BAGASSE FOR AN EVENTUAL PRODUCTION OF BIOETHANOL

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Abstract. Bioethanol represents a promising alternative to gasoline, given that fossil fuel reserves are witnessing an important depletion in the last few years. This paper addresses improving sugars extraction from two energy crops, sweet sorghum, and sweet pearl millet, to produce ethanol. The stalks of these C4 plants are indeed rich in fermentable sugars, essentially sucrose, fructose, and glucose. After being finely chopped, the biomass was pressed with a hydraulic press, and the obtained bagasse was pressed again to extract residual sugars. The bagasse was first humidified either with the juice obtained from the first pressing or water (ratio 1:1 w/w). After that, the wetted bagasse was pressed immediately, after 30 minutes, or after 60 minutes. For both crops, results showed that the humidification's duration does not affect the extraction of residual sugars from the bagasse. The use of water was, however, more efficient than recycling the first pressing's juice. Indeed, an additional 33.4 and 29% of the total fermentable sugars were extracted from sweet sorghum and sweet pearl millet bagasse, respectively, when using water.

INTRODUCTION & LITERATURE REVIEW

Worldwide, the bioethanol production has almost quadrupled in the last decade passing from 138 million barrels in 2002 to 537 million barrels in 2012. The USA and Brazil are the two main producers with 87% of the total production exclusively from maize and sugar cane biomass, respectively (EIA, 2015; Koçar & Civaş, 2013). Some other crops such as wheat and barley are used in Canada (CRFA, 2010). However, the use of these food-crops to produce biofuels has raised many controversies regarding the increase of crop prices and the competition for arable lands (Bouchard, Vanasse, Seguin & Bélanger, 2011). As a result, many studies were carried out to produce bioethanol from alternative energy crops such as sweet sorghum and sweet pearl millet, two annual C4 plants with stalks rich in soluble sugars and having interesting properties. Sweet sorghum (Sorghum bicolor) is a grass originated from Africa (Teetor et al., 2011). It is worldwide grown since it has relatively lower cultural requirements than other crops. Coble, Egg and Schmulevich (1984) indicated that sweet sorghum can be grown in temperate and tropical climatic areas, unlike sugar cane which is a tropical crop. It is also well adapted to different marginal soil types (Zhao et al., 2009) and requires lower quantities of water and fertilizers compared with sugar cane and sugar beet (Basavaraj et al., 2013). According to Billa, Koullas, Monties and Koukios (1997) sweet sorghum contains about 58% of soluble sugars on a dry basis. Sweet pearl millet (Pennisetum glaucum) is the most grown millet specie in Africa (Taylor, 2004). This cereal has an exceptional ability to tolerate drought since it is well adapted to areas considered too dry for sorghum and maize. It also grows well in sandy soils with low fertility (Gulia, Wilson, Carter & Singh, 2007). Andrews and Kumar (1992) showed that sweet pearl millet contains 69% of carbohydrates.

Many studies have focused on either improving the biomass and fermentable sugars yields of sweet sorghum and sweet pearl millet (Teetor et al., 2011; Bouchard et al., 2011; Leblanc, Vanasse, Bélanger and Seguin, 2012) or improving the fermentation process and bagasse valorization as livestock feed or for second generation bioethanol production. Currently, very little information related to sweet sorghum pressing process improvement is available while those concerning sweet pearl millet are almost inexistent. According to Coble et al. (1984) three roll mills used by Bryan, Monroe and Gascho (1981) allowed extracting 40% of sugar from sweet sorghum biomass. However, the sugar extraction yield reached 87% when using a se0ries of tandem roller mills with countercurrent juice flow. With a screw press, Badalov (2008) suggested humidifying the sweet sorghum bagasse with water (ratio 1:1 w/w) to extract about 95% of sugars.

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The main objectives of this study were to investigate the effects of bagasse humidification mode (first pressing juice vs. water) and duration (0, 30 or 60 minutes) on sugar extraction using a hydraulic press.

MATERIALS AND METHODS

Experimental Design

Sweet sorghum (hybrid BMR) and sweet pearl millet (hybrid CSSPM 7) used in this study were seeded on June 11, 2014 at the Université Laval experimental station in Saint-Augustin-de-Desmaures, Quebec, Canada. A seed rate of 10 kg ha^{-1} was considered. Also, 80 kg ha^{-1} of N (40 kg ha^{-1} just before sowing and 40 kg ha^{-1} at the stage of five leaves), 40 kg ha^{-1} of phosphorus, and 40 kg ha^{-1} of potassium were applied just before sowing. The crops were harvested on 8 and 9 September 2014 using a sickle bar mower (New Holland model 451). Two factors were considered in this experiment: bagasse humidification mode (first pressing juice or water) and humidification duration (0, 30, or 60 min). A split-plot design with the humidification mode as main plot was used. Each experiment was replicated three times.

Pressing

After harvesting, the biomass of each crop was finely chopped (5 to 15 mm) with a corn chopper from John Deere and placed into six plastic containers. The content of each container was weighed and then pressed with a hydraulic press designed and built at the Department of Soils and Agri-Food Engineering of Université Laval. The extracted juice volume was measured using a 2000-mL graduated cylinder and the bagasse was retrieved in a container and weighed. Thereafter, the bagasse was humidified either with the first pressing juice or with water at a ratio 1:1 (w/w). The wetted bagasse was then pressed a second time immediately, after 30 min or after 60 min. The second pressing extracted juice was collected and its volume was measured and the bagasse was also weighed.

Biomass Samples

Two samples of 250 g of chopped biomass were taken from each container before the first pressing and were placed into cryovac bags. These bags were then weighed and put in a forced-air oven at 55°C to a constant mass. The dried biomass was re-weighed to determine moister content of the biomass and then the dry matter (DM) content of each container was determined.

Juice Samples

After every pressing, samples of 70 mL of juice were taken and then frozen for further laboratory analyses. Before carrying the analyses, the samples were thawed, shaken, centrifuged, diluted 20 times with distilled water and then two times with acetonitrile. Thereafter, fructose, glucose, and sucrose were separated and quantified with Ultra-performance liquid chromatography (UPLC). Peak identity and sugar quantity were determined by comparison with standards. The sugar content of each sample of juice was computed in a dry basis (g kg^{-1} DM) and then the percentage of additional sugars (fructose, glucose, sucrose, and total sugars) extracted in the second pressing operation was computed.

Data Analysis

Experimental data of sweet sorghum and sweet pearl millet were separately analyzed since comparing the two crops was not our objective. Statistical analyses were performed at the 5% level of significance using the MIXED procedure of the SAS software program, version 9.3.

RESULTS AND DISCUSSION

The humidification mode had a significant effect on the extracted sugars except sweet sorghum fructose (Table 1). On the other hand, the humidification duration wasn't an important factor for both crops. Also, no interaction between the humidification mode and the duration was noticed.

TABLE 1
Treatment Effects on the Extraction of Residual Sugars from Sweet Sorghum and Sweet Pearl Millet Biomass

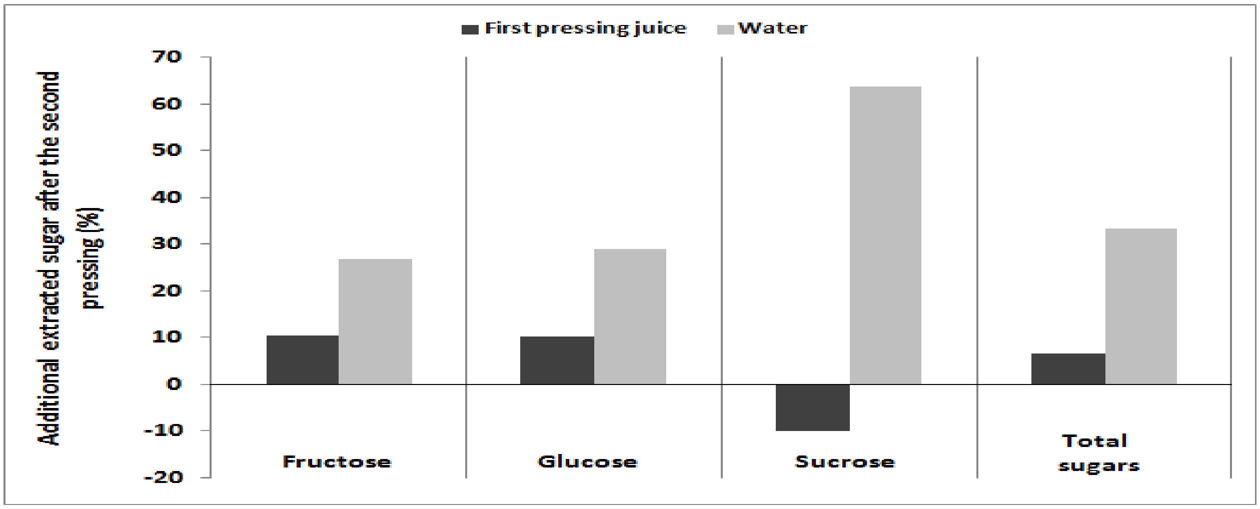
Crop	Source	Fructose	Glucose	Sucrose	Total soluble sugars
Sweet sorghum	Humidification mode (HM)	NS	0.0276	0.0027	0.0077
	Humidification duration (HD)	NS	NS	NS	NS
	HM × HD	NS	NS	NS	NS
Sweet pearl millet	Humidification mode (HM)	<.0001	<.0001	0.0158	0.0158
	Humidification duration (HD)	NS	NS	NS	NS
	HM × HD	NS	NS	NS	NS

NS: not significant

For the sweet sorghum, the extraction of residual fructose, glucose, and sucrose was better when the bagasse was wetted with water than with the first pressing juice (Fig. 1). Indeed, approximately 27% and 29% additional fructose and glucose, respectively, were extracted following a humidification of the bagasse with water against only 10.5% and 10% additional fructose and glucose, respectively, when the bagasse was humidified with the first pressing juice. Regarding the sucrose, the use of water as a wetting solution resulted in better extraction (63.6% more sucrose). However, a 10.2% loss of this sugar was observed when the bagasse was wetted with the first pressing juice.

FIGURE 1

Rate of Additional Sugars Extracted from Sweet Sorghum Bagasse Moistened Either with first Pressing Juice or with Water.

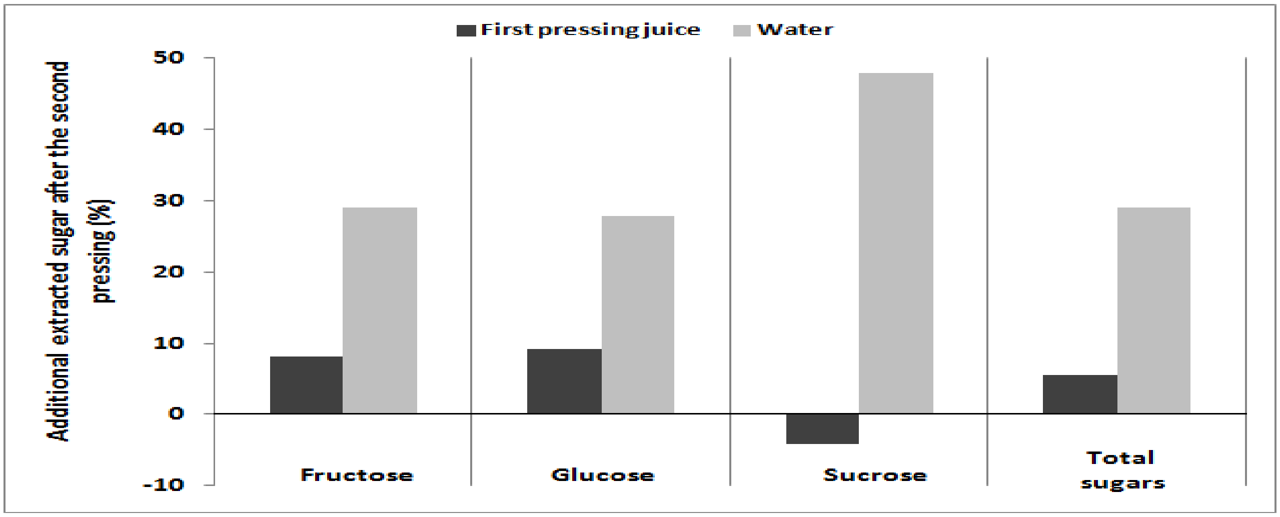


Overall, the same trend was observed for total soluble sugars as about five times more additional sugars were extracted when using water than first pressing juice. Indeed, an average of 6.5% of additional total sugars was obtained when the bagasse was wetted with the first pressing juice while the humidification with water provided an extra 33.4%.

Like sweet sorghum, wetting the bagasse of sweet pearl millet with water was more efficient in extracting residual sugars then using the first pressing juice (Fig. 2). Only 8.1% and 9.2% additional fructose and glucose, respectively, were extracted when the first pressing juice was used as a wetting solution while these rates averaged 29.1% and 27.9% for fructose and glucose, respectively, when the bagasse was wetted with water. Regarding the sucrose, losses of about 4.1% were observed when recycling the first pressing juice while additional 47.9% of that sugar were extracted when the bagasse was wetted with water.

FIGURE 2

Rate of Additional Sugars Extracted from Sweet Pearl Millet Bagasse Moistened Either with First Pressing Juice or with Water



In terms of total soluble sugars, wetting the bagasse with water allowed extracting about five times more additional sugars than when the bagasse was wetted with the first pressing juice. Indeed, about 29% of residual sugars were extracted using water against only 5.6% using the first pressing juice.

Based on the results obtained for sweet sorghum and sweet pearl millet, it is clear that wetting the bagasse with water is more efficient in extracting residual sugars compared to the use of the first pressing juice. Since the chopped biomass of sweet sorghum and sweet pearl millet is rich in fibers which are insoluble carbohydrates (cellulose, hemicellulose, and lignin), a significant amount of juice rich in sugars remains retained by the fibers forming the residual sugars in the bagasse following a single pressing. This has been demonstrated by Bryan et al. (1985) who found a negative linear correlation between the juice yield and the content of sweet sorghum in fibers.

When using the first pressing juice, the low rate of additional sugars extraction could be explained by the high concentration of the sugars in the juice. When this latter enters in contact with the bagasse, sugars exchange between the bagasse and the juice is therefore limited. More specifically, the loss of sucrose for both crops when recycling the first pressing juice may be due to the action of invertase, an enzyme responsible for the hydrolysis of sucrose into its two monomers glucose and fructose. As mentioned by Sturm (1999) this enzyme is present in most plant species in different isoforms in the cell wall, in the cytosol, and in the vacuoles. Mutual action of these invertase isoforms, in the presence of other regulators, helps maintaining a certain concentration of sucrose in the cells since it has a role in cell differentiation. When the sorghum or millet plant is harvested, chopped, and pressed, the control of invertase action process is lost. Being in contact with the sucrose, the invertase starts therefore exercising its hydrolytic action in the juice which explains the loss of sucrose. Even if wetting the bagasse with this juice can help extracting some amount of residual sucrose, the action of invertase appears to be more important. Therefore, the increase of glucose and fructose rates may be explained by the hydrolysis of sucrose and the extraction of residual sugars from the bagasse.

Regarding the use of water to wet the bagasse, the rate of obtained additional total sugars was five times higher than that obtained using the first pressing juice. Indeed, added water allows leaching the bagasse fibers by extracting residual sugars by diffusion resulting therefore in greater residual sugars extraction.

CONCLUSION

Wetting the bagasse had a significant effect on residual sugars extraction from sweet sorghum and sweet pearl millet biomass. The use of water to wet the bagasse was however more efficient than the use of first pressing juice. On the other hand, wetting duration had no significant effect on sugars extraction.

A second pressing of sweet sorghum and sweet pearl millet bagasse with the addition of water represents an important step in the bioethanol production process since it allows extracting more residual sugars. However, more research is needed to optimize the water:bagasse ratio.

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