

STUDY OF THE CHEMICAL COMPOSITION OF SWEET SORGHUM STALKS DEPLETED IN CARBOHYDRATES WITH APPLICATIONS IN OBTAINING BIOETHANOL

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Abstract: Sweet sorghum is a great energy crop that shows the benefits to ecosystems, energy and economics, being a valuable source of energy of the category 1st, 2nd and 3rd. bioethanol generation.

Purpose of the paper is to study the chemical composition of sweet sorghum stalks depleted in carbohydrates with applications in obtaining ethanol.

It shows appreciable compositional values of free sugars, starch, cellulose, hemicellulose and lignin.

All these components can be easily made available as fermentable carbohydrates leading to the production of products with high economic value (bioethanol).

INTRODUCTION

The quantities of non-renewable (conventional) energy resources are limited and they have a considerable negative environment impact e.g. increased greenhouse gas emissions. Therefore, one of the challenges for the society is to meet the growing demand for energy for transportation, heating and industrial processes; also to provide raw materials for the industry in a sustainable way and to reduce greenhouse gas emissions. Our energy systems will need to be renewable and sustainable, efficient and cost-effective, convenient and safe [Mojović *et al.*, 2009; Jouve, 2006; Blanch *et al.*, 2008].

Bioethanol produced from renewable biomass, such as sugar, starch, or lignocellulosic materials, is one of the alternative energy resources, which is both renewable and environmentally friendly. Although, the priority in global future ethanol production is put on lignocellulosic processing, which is considered as one of the most promising second-generation biofuel technologies, the utilization of lignocellulosic material for fuel ethanol is still under improvement [Mojolovic *et al.*, 2009].

The varied raw materials used in the production of ethanol via fermentation are conveniently classified into three main types of raw materials: sugars, starches, and cellulose materials. Sugars (from sugarcane, sugar beets, sweet sorghum, molasses, and fruits) can be converted into ethanol directly. Starches (from corn, cassava, potatoes, and root crops) must be hydrolyzed to fermentable sugars by the action of enzymes from malt or molds. Cellulose (from wood, agricultural residues, waste liquor from pulp, and paper mills) must likewise be converted into sugars, generally by the action of mineral acids. Once simple sugars are formed, enzymes from microorganisms can readily ferment them to ethanol [Liu and Shen, 2008]. The progress possibilities are discussed in the domain of feedstock choice and pretreatment, optimization of fermentation, process integration and utilization of the process byproducts [Mojolovic *et al.*, 2009].

Large possibilities exist for biomass resources (in particular energy crops) to penetrate the power generation and the transport markets: however, the demonstration of the economic viability of this new activity is essential for its large deployment. The feasibility of Sweet Sorghum cultivation for renewable and sustainable production of transport fuel (bioethanol, and - in case - hydrogen and methanol), energy (electricity and heat) and other products (as animal feed, pulp for paper, charcoal, activated coal) with commercially available technologies is necessary for tested [Chiaromonti *et al.*, 2002].

When grasses availability is limited due to scarcity or water shortage in dry periods the agricultural residues, with the majority of it comes from irrigated scheme as well as rain fed areas, emerges as important alternative source of feed for ruminant. However, sorghum stover is the most abundant cereal residues. With respect to animal feeding the major biological constraints for using crop residues are the low protein content and the vast amount of lignocellulosic

material which has a very low digestibility. Techniques to improve the feeding value of crop residues by physical, chemical or biological means have not been adopted widely by small farmers in developing countries mainly due to economical constraints [Fadel Elseed *et al.*, 2007].

The weak relationship between grain yield and quality characters of cereal crop residues provides a potential exists for selecting or breeding varieties with improved straw and stover value without sacrificing grain production. Therefore, genetic enhancement may provide an alternative and practical strategy for improvement of nutritive value in cereal straws and stovers. The objective of this study is to evaluate the nutritionists and crop breeders [Fadel Elseed *et al.*, 2007]

Sweet sorghum (*Sorghum bicolor*) is similar to common grain sorghum with a sugar-rich stalk. Sweet sorghum is characterized by wide adaptability, drought resistance, water logging tolerance, saline-alkali tolerance, rapid growth, high sugar accumulation, and biomass [Reddy and Reddy, 2003].

Variability has been recorded in sweet sorghum for grain yield from 1.5 to 7.5 t ha⁻¹, ranging from 13 to 24%, for sucrose from 7.2% to 15.5%, for stalk yield from 24 to 120 t ha⁻¹ and for biomass yield from 36 to 140 t ha⁻¹. Sweet sorghum has a biomass product ion capacity equal or superior to sugarcane in the tropics. Alcohol is produced at 6106 L ha⁻¹ from sweet sorghum while only 4680 L ha⁻¹ from sugarcane is produced. There are several advantages of using sweet sorghum instead of sugarcane for alcohol product ion. These are: sweet sorghum is harvested in four months (whereas, the first cut of sugarcane is 18 months after planting); sweet sorghum product ion can be completely mechanized; the crop can be established from seed; the grain may be used as either food or feed; the stillage from sweet sorghum has a higher biological value than the bagasse from sugarcane when used as forage for animals. Stillage obtained after extract ion of juice from the stalks of sweet sorghum contains similar levels of cellulose as sugarcane bagasse; therefore, it has a good prospect as a raw material for pulp product. It could be processed as a feed for ruminant animals. Furthermore, it is rich in micronutrient s and minerals. It reported that jaggery prepared from sweet sorghum juice contained 78.1% sucrose and 8.8% reducing sugars while that from sugarcane contained 84.2% sucrose and 7.5% reducing sugars [Reddy and Reddy, 2003].

This study focuses on the dry weight, carbohydrate, and fiber content of enhanced sweet sorghum varieties LC99, Prut, 11100, 11061, comparatively with Sudangrass, wild type.

MATERIALS AND METHODS

The biomass used in the experiment contains in residual straw of sweet sorghum LC99, Prut, 11100, 11061, Sudangrass varieties from the collection of the Cereal and Technical Plants Research Institute I.C.C.P.T. - Fundulea, Călărași. All reagents are analytically pure. Right after harvesting samples are dried at 50°C until constant weight [Spigno *et al.*, 2008; Faithull, 2002; AOAC, 1995]. Free reducing sugars are extracted by boiling and then solid residues are dried at 50°C until constant weight. Dried residual sorghum straw have been chopped and sifted through a 3 x 3 mm porosity sieve.

All samples have been worked three times. For further analysis water extract was prepared according to Spingo *et al.*, 2008. For determining free reducing glucids DNS was used [Mandels and colab., 1976], glucids hydrolysis for total glucids determination was conducted according to Petrescu and colab., 1967. Pentose quantification was made with orcinol in acid environment [Iordachescu and Dumitru, 1982]. Sugar content determination was conducted according to Halhoul and Kleinberg method, 1972. Polifenols determination after Hiuneburg and colab. method, 2006, requires Folin-Ciocalteu reagent

From the biomass the next analysis can be made: residues after complete burn, organic substance [Petrescu and colab., 1967; Faithfull, 2002; AOAC, 1995], total nitrogen and total protein through Kjeldal method [Petrescu and colab., 1967; Iordachescu and Dumitru, 1982], amidon with potassium iodide in acid environment [du Boil and Schaffler, 1974], NDF with SDS [Faithull, 2002], ADF with cetazol replacing CTAB with similar results [Faithull, 2002].

RESULTS AND DISCUSSIONS

The chemical compositions of four improved varieties compared to wild type Sudangrass.

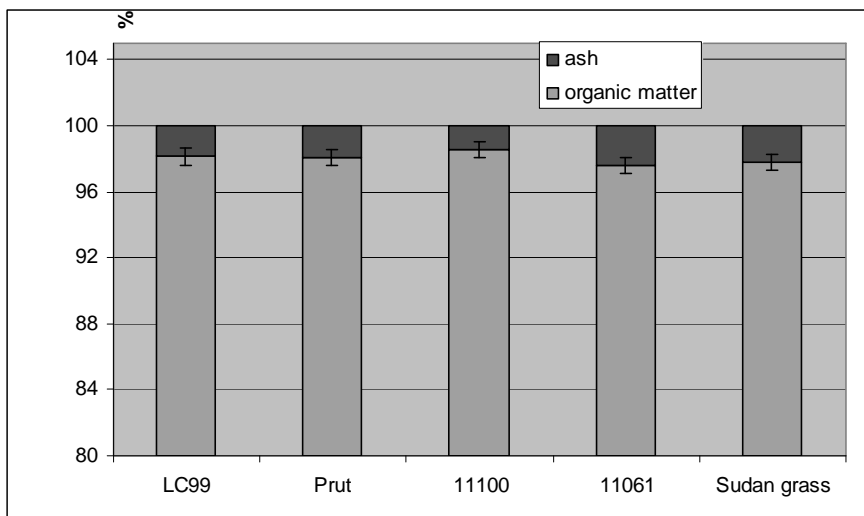


Figure 1. The ash/organic substance ratio (%) of stover fractions of five stem sorghum varieties.

It is visible that 11061 varieties have the largest ash quantity and 11100 the smallest. Fadel Elseed *et al.* study from 2007 presents the following data.

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18					
Organic matter (%)	95.2	92.4	90.4	93.3	93.4

In comparison to the results of this study the ash ratio is maximum 3%, which indicates that organic matter available for bioethanol is at least 97%.

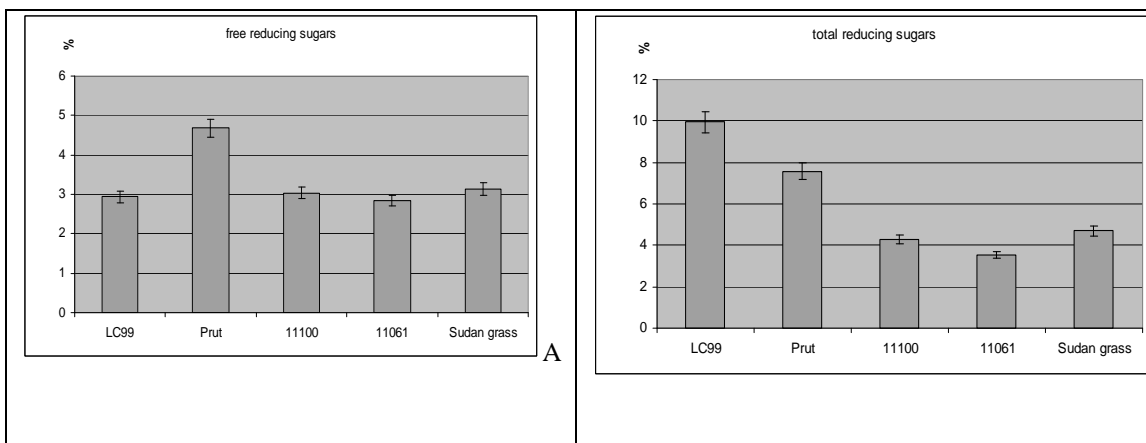


Figure 2. Reducing glucides content (%): A) free reducing glucides B) total reducing glucides

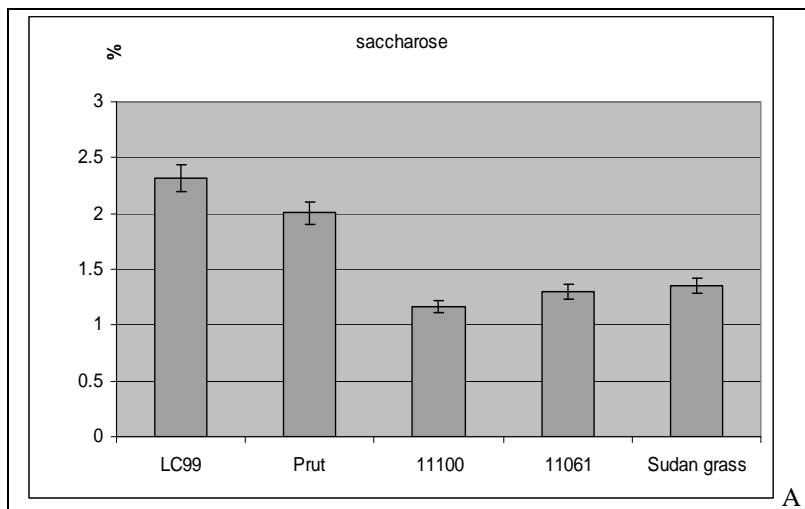
The content in reducing glucides of residual sweet sorghum straw.

It is visible that the content in reducing glucides of residual sweet sorghum straw is higher with less than 2% in Prut varieties compared to the other samples. The lowest glucid content was almost 3% in 11061 varieties. The medium value was around 3% (fig. 2A) Ricaud *et al.*, in 1979 showed that fresh sorghum straw have a free reducing sugar content between 13.2 and 15.6%. Channappagoudar *et al.*, 2007, publish a study showing that fresh sorghum straw have a free reducing sugar content between 0.130 and 0.711 g/100g sample.

The total glucid content of the residual sweet sorghum straw has been observed to have increased with few under 2 percents in LC99 and Prut varieties in comparison to the rest of the samples. The lowest total glucid content was found in 11061 varieties proportionally to the free sugar content (fig. 2B).

Bennett and Anex, in 2009, say that fresh sorghum straw have total reducing sugar content between 10.5-13.9%.

Also, Channappagoudar *et al.*, in 2007, present that ca fresh sorghum straw have a total reducing sugar content between 0.264-1.967 g/100g sample.



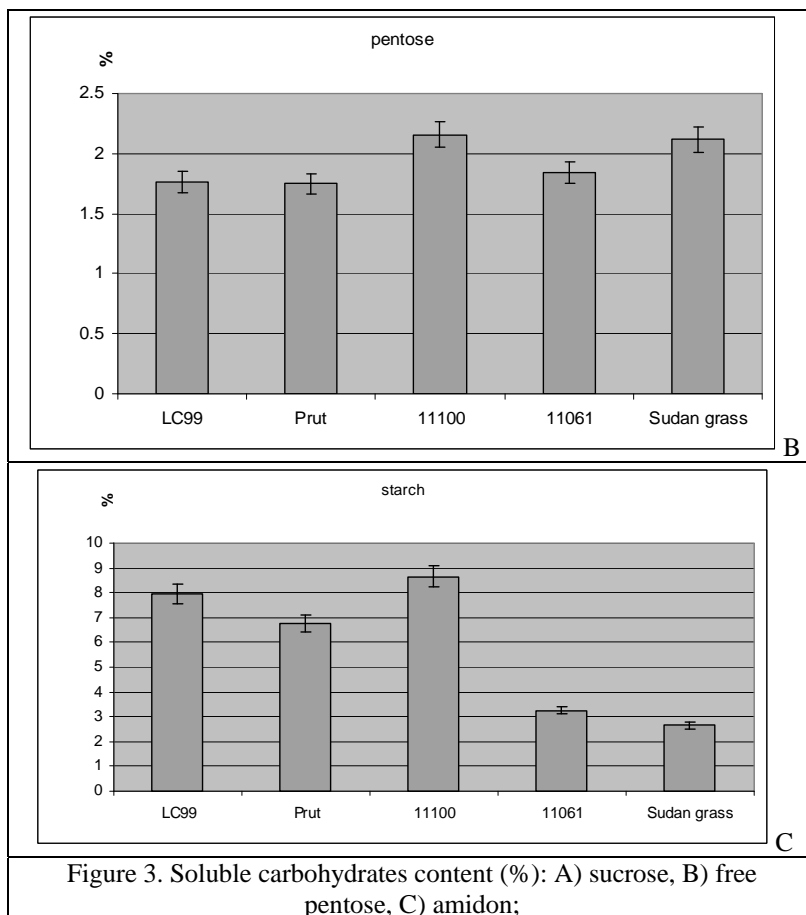


Figure 3. Soluble carbohydrates content (%): A) sucrose, B) free pentose, C) amidon;

For the soluble carbohydrates content (%), the values are as follows: for sucrose is high for LC99 and Prut varieties, in correlation to the free and total glucides content; the lowest is for 1110 (fig. 3A); for free pentose is high for 11100 and Sudangrass, in correlation to the total and free glucides content; the lowest is for LC99 and Prut varieties (fig. 3B); for amidon is high in 11100 and LC99 varieties; the lowest is for 11061 and Sudangrass (fig. 3C).

Ricaud *et al.*, 1979, have studied sweet sorghum straw with sucrose content between 8.1 and 13%. Also, they had free pentose content between 1,1-1,9%.

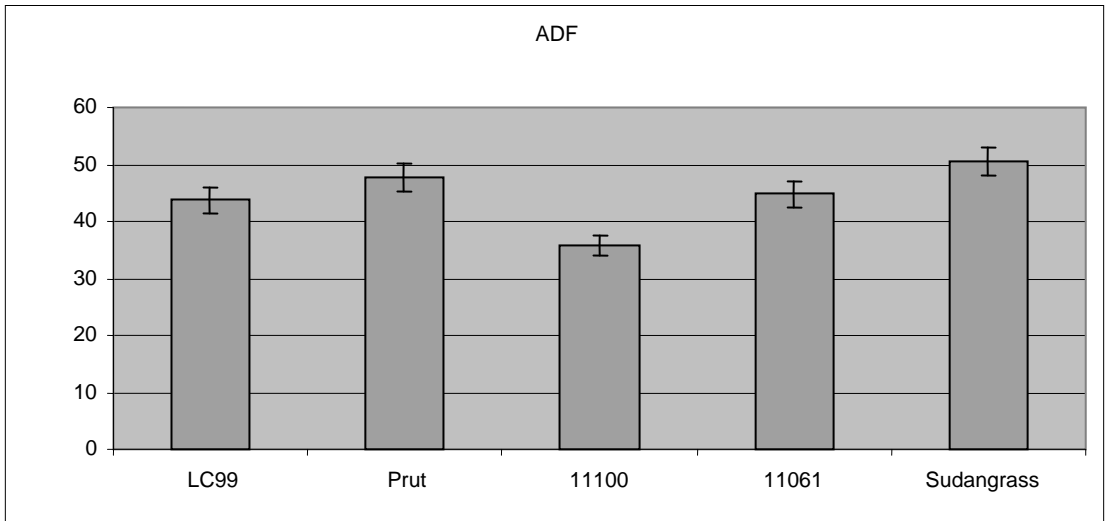


Figure 4. ADF content.

The highest ADF content is for Sudangrass, the lowest being for 11100 varieties (fig. 4).

Fadel Elseed *et al.*, 2007, show that ADF content is comprised between 45-60%, depending on the varieties.

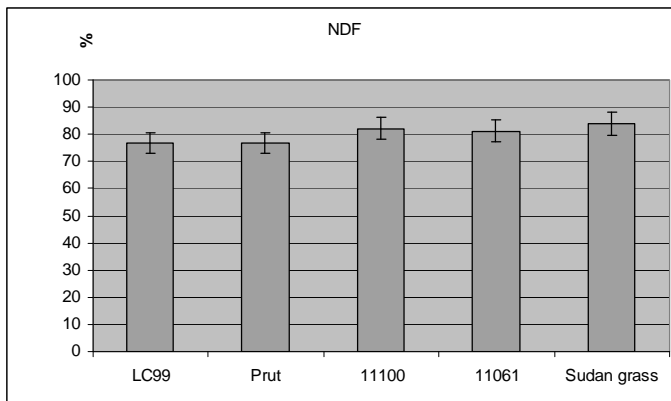


Figure 5. NDF content.

The highest NDF content is for Sudan grass, the lowest being for LC99 and Prut (fig. 5).

Fadel Elseed *et al.*, 2007, show that NDF content (%) is comprised between 60.6 and 78, depending on the varieties.

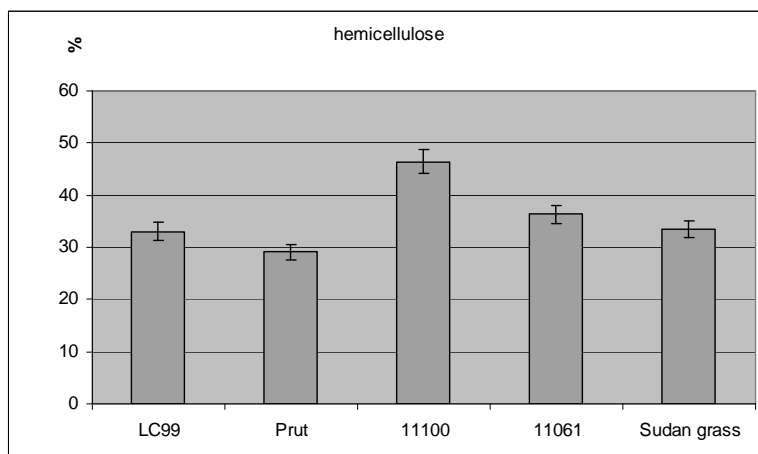


Figure 6. Hemicellulose content.

The hemicellulose content is highest for 11100 and the lowest for Prut varieties (fig. 6). Fadel Elseed *et al.*, 2007, show that hemicellulose has values between 4.2 and 18.5%.

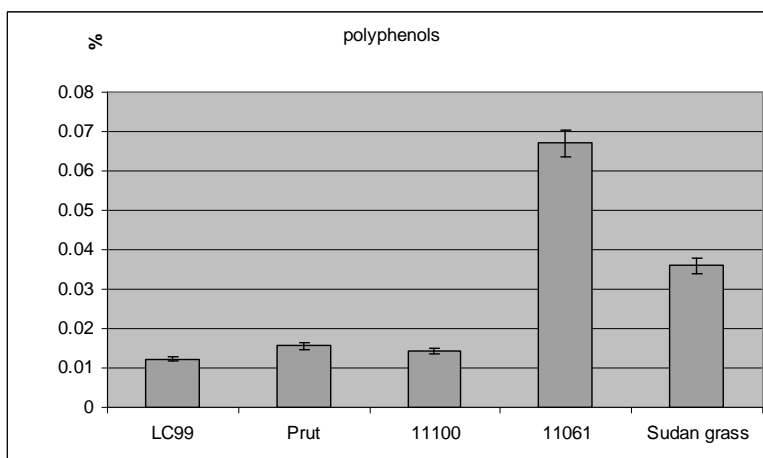


Figure 7. Polifenols content.

According to figure 7 polyphenols have the highest value in 11061 varieties, followed by Sudangrass, the rest of the samples having significantly lower values. Dicko *et al.*, 2006, showed that untreated sorghum straw have a polyphenols content between 0.5 and 3%. Dykes and Rooney, 2006, found that untreated sorghum straw has polyphenols content between 0.10 and 0.59 mg catechin equivalent /100 mg sample.

Among cereals, sorghum has the highest content of phenolic compounds reaching up to 6% (w/w) in some varieties. Contents of phenolic compounds and phenolic oxidizing enzymes are strongly associated with food quality. Enzymes involved in the biosynthesis and oxidation of

phenolic compounds have been shown to be determinants for the quality of plant-derived foods. The presence of PAL activity in sorghum grain and its activation upon germination was assessed. Although PAL is indirectly involved in the synthesis of almost all phenolic compounds, its activity was not correlated with the contents in phenolic compounds in both ungerminated and germinated varieties. This lack of correlation may be due to the presence of phenolic oxidizing enzymes in the grain [Dicko *et al.*, 2006].

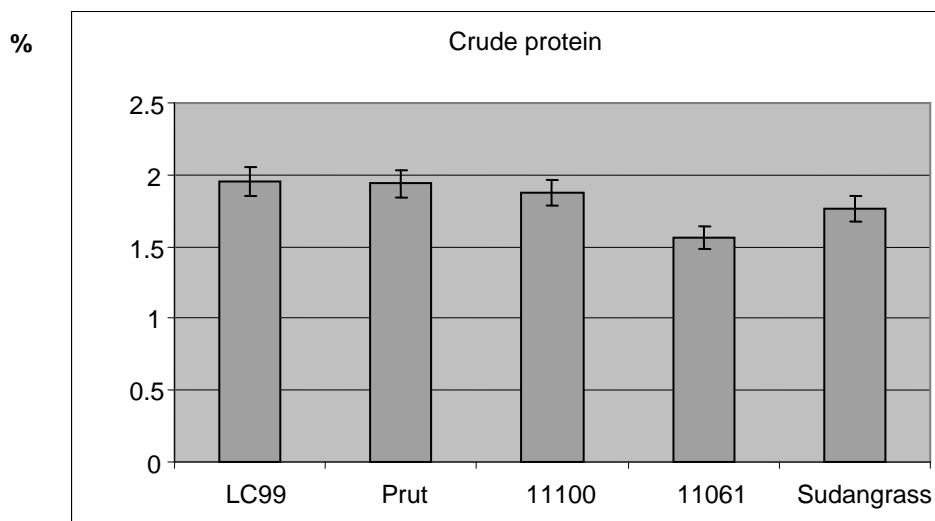


Figure 8. Total protein content.

The total protein quantity is high for LC99 and Prut and very low for 11061 (fig. 8). Fadel Elseed *et al.*, 2007, show that the protein content is between 3.2 and 7.4.

CONCLUSIONS

Studies of chemical composition of sweet sorghum in the perspective of choosing improved varieties for optimal bioethanol production present interesting data. Each of the analyzed sorghum varieties has variable proportions of glucides, amidon and fibers. It has been observed that LC99 and Prut varieties have a higher free and total glucides quantity, 11100 varieties has a higher amidon quantity and 11061 and Sudangrass have a higher content of fibers.

REFERENCES

Chiaromonti D., A. Agterberg, G. Grassi, H.-P. Grimm, B. Coda, (2002), "Large Bio-Ethanol Project from sweet sorghum in China and Italy (ECHIT): Description of site, process schemes and main products", 12th European

- Conference on Biomass for Energy, Industry and Climate Protection*, 17-21 June 2002, Amsterdam, The Netherlands 1114-1117;357 [Chiaromonti *et al.*, 2002]
- Bennett A. and R. Anex, (2009), “Production, transportation and milling costs of sweet sorghum as a feedstock for centralized bioethanol production in the upper Midwest”, *Bioresource Technology*, 100:1595-1607; 386 [Bennett and Anex, 2009]
- Fadel Elseed A.M.A., N. Nor Eldaim and E.O. Amasaib, (2007), “Chemical Composition and *in Situ* Dry Matter Degradability of Stover Fractions of Five Sorghum Varieties” *Journal of Applied Sciences Research*, 3(10):1141-1145; 728 Fadel Elseed *et al.*, 2007]
- Association of Official Agricultural Chemists (AOAC), (1995), „Official Methods Of Analysis of AOAC International”, 16th Edition, Supplement, Virginia, USA; AOAC, 1995]
- Blanch H., P. Adams, K. Andrews-Cramer, W. Frommer, B. Simmons and J. Keasling, (2008), “Addressing the need for alternative transportation fuels: The joint BioEnergy Institute”, *ACS Chemical Biology*, 3(1):17-20; Blanch *et al.*, 2008].
- Bogo A., Mantle P.G. and Harthmann O.E.L., (2004), “Screening of sweet sorghum accessions for inhibition of secondary sporulation and saccharide measurements in honeydew of *Claviceps Africana*”. *Fitopatologia Brasileira* 29:086-090; 458 [Bogo *et al.*, 2004]
- Channappagoudar B. B., N. R. Biradar, J. B. Patil and S. M. Hiremath, (2007), “Assessment of Sweet Sorghum Genotypes for Cane Yield, Juice Characters and Sugar Levels”, *Karnataka J. Agric. Sci.*,20(2):294-296; 460 [Channappagoudar *et al.*, 2007]
- Dicko M., H. Gruppen, A. Traoré, A. Voragen and W. van Berkel, (2006), *Review* “Phenolic compounds and related enzymes as determinants of sorghum for food use”, *Biotechnology and Molecular Biology Review*, 1(1):21-38; 456 [Dicko *et al.*, 2006]
- Du Boil and Schaffler, (1974), „A simplified starch determination for raw sugars”, *Proceedings of The South African Sugars Technologists’ Association – April 1974* p39-41; du Boil and Schaffler, 1974
- Dykes L. and L. Rooney, (2006), *Review* “Sorghum and millet phenols and antioxidants”, *Journal of Cereal Science*, 44:236-251; 457 Dykes L. and L. Rooney (2006)
- Faithull N.T., (2002), „Methods in agricultural chemical analysis: a practical handbook”, Chapter 7: „The analysis of animal feed and plant material” pg. 124-153; Faithull, 2002;
- Halhoul M.N. and Kleinberg I., (1972), „Differential determination of glucose and fructose, and glucose- and fructose- yielding substances with anthrone”, *Analytical Biochemistry*, 50:337-343
- Hiunenburg I., Dormann H. J. D. and Hiltunen R., (2006) “Antioxidant activities of extracts from selected culinary herbs and species” *Food Chemistry* 97:122-129; Hiunenburg and colab., (2006)
- Iordachescu and Dumitru, (1982), „Biochimie practică, partea a-II-a: Aminoacizi, glucide și lipide”, Universitatea din București; Iordachescu și Dumitru, 1982
- Jouve C., (2006), Division for Science and Process Engineering of Agricultural Products, Biosystems and process engineering, Joint Research Unit, INRA-CNRS-INSA Toulouse, Bioenergies, *Report 2003-2006*; Jouve, 2006;
- Liu R., F. Shen, (2008), “Impacts of main factors on bioethanol fermentation from stalk juice of sweet sorghum by immobilized *Saccharomyces cerevisiae* (CICC 1308)”, *Bioresource Technology*, 99:847-854; 1091 [Liu and Shen, 2008]
- Mandels M., Andreotti R and Roche C., (1976), „Measurement of saccharifying cellulase”, *Biotechnol. Bioenerg. Symp.* 6:21-33; Mandels and colab., 1976
- Mojolovic L., Pejcin D., Grujic O., Markov S., Pejcin J., Rakin M., Vukasinovic M., Nikolic S., Savic D., (2009), “Progress in the production of bioethanol on starch based feedstocks”, *Chemical Industry & Chemical Engineering Quarterly* 15(4):211-226; 1085 [Mojolovic *et al.*, 2009]
- Mojovic L., D. Pejcin, O. Grujic, S. Markov, J. Pejcin, M. Rakin, M. Vukasinovic, S. Nikolic, D. Savic, (2009), “Progress in the production of bioethanol on starch-based feedstocks”, *Chemical Industry & Chemical Engineering Quarterly*, 15(4):211-226; Mojovic *et al.*, 2009;
- Petrescu C., Tascenco V., F. Saghin, I. Feredan, I. Lupan, (1966), „Analiza tehnică a nutrețurilor” Întreprinderea Poligrafică „Tiparul”, București; Petrescu and colab., 1967
- Reddy BVS and S. Reddy, (2003), “Sweet Sorghum: Characteristics and Potential”, *ISMN*, 44:26-28; 1087 [Reddy and Reddy, 2003]

Ricaud R., B. Cochran, A. Arceneaux and G. Newton, (1979), "Sweet sorghum for sugar and biomass production in Louisiana",
<http://74.53.10.34/~ssea2008/SS%20Publications/USDA/USDA%20SS%20Report%201979.pdf> [Ricaud *et al.*, 1979] 295

Spigno G., T. Pizzorno, D. M. De Faveri, (2008), "Cellulose and hemicelluloses recovery from grape stalks", *Bioresource Technology*, 99:4329-4337; Spigno and colab., 2008

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