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Effect Of Agromorphological Diversity and Botanical Race on Biochemical Composition in Sweet Grains Sorghum [*Sorghum bicolor* (L.) Moench] of Burkina Faso

ABSTRACT

Sorghum bicolor (L.) Moench is an under-harvested crop in Burkina Faso. It is grown mainly for its sweet grains in the pasty stage. However, the precocity of the cycle and the sweet grains at pasty stage make it an interesting plant with agro-alimentary potential during the lean season. This study was carried out to identify the main sugars responsible for the sweetness of the grains at the pasty stage and their variation according to the agro-morphological group and the botanical race. Thus, the grains harvested at the pasty stage of fifteen (15) accessions selected according to the agro-morphological group and botanical race were lyophilized and analyzed by High Performance Liquid Chromatography (HPLC). The results reveal the presence of four (4) main carbohydrates at the pasty stage of grains such as fructose, glucose, sucrose and starch. Analysis of variance revealed that these carbohydrates discriminate significantly the agro-morphological groups and the botanical races. Moreover, with exception of the sucrose, the coefficient of determination (R^2) values shows that the agro-morphological group factor has a greater effect on the expression of glucose, fructose and starch than the botanical race. Group III and *caudatum* race have the highest levels of fructose and would be the sweetest. While group IV and the *guinea-bicolor* race with the low value of fructose would be the least sweet. Fructose is therefore, the main sugar responsible for the sweetness of the pasty grains of sweet grains *sorghum*.

Keywords: *Sorghum*, neglected culture, *sorghum race*, biochemical composition, Burkina Faso

Introduction

Very poorly known in the world, sweet grains sorghum (*Sorghum bicolor* L. Moench) is one of the less valued types of sorghum. So far, Burkina Faso Ministry of Agriculture doesn't take this sorghum into account in the national agricultural statistics on sorghum. Then, information on national production and the extent of its cultivation are not available. Sweet grains sorghum is an under-harvested crop that is produced mainly around the dwelling houses and generally associated with other cereal crops (maize, millet, and other types of sorghum) or legumes (Sawadogo et al., 2014a). Its sweet grains in the pasty stage are eaten directly

by mastication. Their harvesting usually takes place before the main food crops. It constitutes a welding food in rural areas (Nebié et al., 2012). The sale of panicles harvested at the pasty grains stage generates income for producers and retailers. In addition, its leaves and stem are exploited as fodder or as domestic fuel (Sawadogo et al., 2014a). Compared to common sorghum and sweet sorghum, sweet grains sorghum is a minor plant in the Burkina Faso agrarian system. The cultivation of this sorghum is increasingly abandoned. This seriously threatens the preservation of its genetic resources. Yet now, all actors of plant breeding are unanimous on the need to preserve the genetic resources of plants. They constitute a national and universal heritage that can be used as direct sources of adapted varieties, or as

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sources of important traits for improvement or as basic material for phylogeny studies (Amri et al., 1997).

The different genetic diversity carried out on sweet grains sorghum have focused on the agro-morphological and racial diversity of accessions collected in the north center (Nebié et al., 2012), the north (Sawadogo et al., 2014b) and all agro-climatic zones of Burkina Faso (Sawadogo et al., 2014a). These studies highlighted the existence of diversity within this sorghum and the possibilities of improvement by direct selection. They also showed the predominance of the main race *caudatum* and intermediate *caudatum-guinea* (Sawadogo, 2015). However, the biochemical composition of this sorghum, in particular the identification of the main sugars responsible for its sweet taste has not yet been achieved.

The objective of this study is to contribute to a better knowledge of the biochemical diversity, in particular, determination of the carbohydrate composition of the pasty grains of the sweet grains sorghum of Burkina Faso. This involves identifying the main sugars responsible for the sweet taste of the grain in the pasty state, quantifying them, determining the variation in the content of the main sugars according to the agro-morphological groups and the botanical races.

Materials and Methods

Plant material

The plant material consists of freeze-dried grains of

fifteen (15) sweet grains sorghum accessions selected in the sorghum gene bank of the University of Ouagadougou (Burkina Faso). The grains were harvested at the pasty stage where the sweet flavor is highest. The choice of these accessions was made on the basis of their own performance in order to integrate the different agro-morphological groups defined and the different botanical races identified (Sawadogo, 2015). The distribution of the fifteen accessions selected according to agro-morphological groups and botanical races is presented in Table 1.

Carbohydrates analysis and quantification

Grains lyophilization

The grains harvested in the pasty stage were previously freeze-dried in the laboratory of virology of Institute of Environment and Agricultural Research (INERA) in Kamboinsé (CREAF).

Quantification of carbohydrates

Main carbohydrates identification and quantification were carried out in the biochemistry laboratory of the Genetic Improvement and Adaptation of Mediterranean and Tropical Plants Unit (AGAP) of the CIRAD Montpellier in 2013. The analysis was concerning all carbohydrates which have concentrations above 1 mg / g DM.

The identification of the main soluble sugars (glucose, fructose, sucrose) was carried out according to the method current used (Mialet-Serra et al., 2005). Quantification was performed by High Performance Liquid chromatography

Table 1. Distribution of the fifteen accessions characterized according to the Agro-morphological groups and botanical races

		Accessions number	Characteristics	Total
Agro-morphological groups	I	02	Long cycle, high grain yield	15
	II	05	Long cycle, Average grain yield	
	III	04	Average cycle, Average grain yield	
	IV	04	Short cycle, Low grain yield	
Botanical races	<i>Caudatum</i>	07	Farinaceous seeds, dissymmetrical seeds	15
	<i>Caudatum-guinea</i>	06	Farinaceous seeds, dissymmetrical seeds, rotation of seeds	
	<i>Guinea-bicolor</i>	02	Rotation of seeds long glumes	

Method (HPLC) with pulsed amperometric detection (Dionex, Salt Lake City, UT, USA).

As for the starch contained in the insoluble fraction of the pellet, it was solubilized with 0.02 N sodium hydroxide at 90 ° C during 90 min and hydrolysed with amyloglucosidase (35 U) in a water bath (50 ° C) for 90 min after evaporation of the 80% ethanol using the Speedvac (60 min). Glucose was quantified using hexokinase and glucose-6-phosphate dehydrogenase at 340 nm (Boehringer, 1984). The results were expressed in milligrams of equivalent glucose per gram of dry matter (DM).

In order to have more information about prices of panicles, an inquiry was carried out near ten retailers located at the edge of the roads.

Statistical analysis of data

The data were analyzed with Statistica version 6 and Xlstat pro 7.1 software. A one-way analysis of variance by considering the factors agro-morphological group and botanical race and a matrix of correlations were carried out with Statistica. In order to determine the variance share of the quantitative variables attributable to the factors botanical race and agro-morphological group, the coefficients of determination (R^2) were established with the software Xlstat pro 7.1. The means separation test of Newman-Keuls at the 5% threshold was carried out with also Xlstat pro 7.1 to identify the carbohydrates that discriminate the botanical races and the agro-morphological groups.

Results

Carbohydrate composition of grains in pasty stage

Variation of carbohydrate composition of accessions

Two monosaccharides (glucose, fructose), one disaccharide (sucrose) and one polysaccharide (starch) were assayed in the freeze-dried paste grains. The results of the carbohydrate content in sweet grains at pasty stage recorded in Table 2 show a variation of the content of the four carbohydrates analyzed. An important amplitude of variation

with a low content. However, the other two sugars, glucose and fructose have intermediate contents.

Variation of the carbohydrate composition according to the agro-morphological group

Comparison of the biochemical composition of grain between agro-morphological groups consigned in Table 3 reveals that glucose, fructose and starch carbohydrates discriminate significantly groups at the thresholds of 1%. Only sucrose content isn't significantly different between groups. The variation of carbohydrates content between the agro-morphological groups (Figure 1) shows that Group III has the highest levels of glucose, fructose and the lowest starch content while group IV has the highest starch content and the lowest contents for the two monosaccharides.

Variation of the carbohydrate composition according to the botanical race

With the exception of the sucrose, the others carbohydrates content discriminate significantly the botanical races (Table 4). A high degree of heterogeneity is observed between the botanical races for the glucose, fructose and sucrose composition. That is revealed by the coefficients of variation higher than 30%. The *caudatum* race has the highest levels of glucose and fructose while the intermediate race *guinea-bicolor* has the highest levels of sucrose and starch (Figure 2).

Comparison of the influence of agro-morphological group factor and botanical race factor on variation in the carbohydrate composition of accessions

The analysis of variance carried out according to the botanical race factor and agro-morphological group shows that the carbohydrate composition of the grains is more dependent on the agro-morphological group than the botanical race. The values of the coefficients of determination (R^2) of the different carbohydrates consigned in Table 5 show that the botanical race with values of R^2 included between 21% and 36.6% has a greater effect than the agro-morphological group in the expression of the grains sucrose

Table 2. Variation of the biochemical composition of the pasty grains of sweet grains sorghum accessions

Glucides	Min.	Max.	Mean
Glucose (mg/g DM)	5.786	55.247	26.067
Fructose (mg/g DM)	6.598	66.097	30.861
Sucrose (mg/g DM)	0.41	8.63	2.899
Starch (mg/g DM)	373.616	688.964	549.047

DM : dry matter; Min. : Minimum ; Max. : Maximum

is observed between the extreme values for the four carbohydrates. Starch is indeed the majority carbohydrate with high values, while sucrose is the minority carbohydrate

content. But, for the other carbohydrates such as glucose, fructose and starch, agro-morphological group factor has the greater effect on their expression.

Correlations between metered sugars

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Table 3. Results of the analysis of variance of the carbohydrate content according to the agro-morphological group factor

	Agro- morphological groups				CV (%)	F
	I	II	III	IV		
Glucose (mg/g DM)	13,62b	34,69a	37,43a	10,15b	54,6	13,56**
Fructose (mg/g DM)	16,09bc	41,39ab	44,36a	11,59c	57,6	12,618**
Sucrose (mg/g DM)	2,281a	2,693a	3,46a	2,907a	94,9	0,367ns
Starch (mg/g DM)	564,65ab	501,42b	495,56b	654,24a	14,8	12,632**

DM : dry matter, CV : Coefficient of Variation, F : Fisher coefficient, ns : not significant at 5 %, ** : significant at 1 %, R² : coefficient of determination, a,b,c: the values followed by the same letters are not significantly different

Table 4. Results of the analysis of variance of the carbohydrate content according to the botanical race factor

	Botanical races			CV (%)	F
	Caudatum	Caudatum- guinea	Guinea- bicolor		
Glucose (mg/g DM)	33,3a	24,28ab	6,12b	63,2	7,609**
Fructose (mg/g DM)	40,47a	27,68ab	6,76b	64,9	8,166**
Sucrose (mg/g DM)	3,221ab	1,786b	5,116a	81,8	5,596**
Starch (mg/g DM)	501,28b	561,14b	679,95a	15,7	11,989**

DM : Dry matter, CV : Coefficient of Variation, F : Fisher coefficient, ** : significant at 1 %, R² : coefficient of determination, a,b,c: the values followed by the same letters are not significantly different.

Table 5. Coefficients of determination (R²) of the different carbohydrates according to the agro-morphological group and the botanical race

	Agro-morphological groups (R ² %)	Botanical race (R ² %)
Glucose (mg/g DM)	49.8	26.6
Fructose (mg/g DM)	48	28
Sucrose (mg/g DM)	2.6	21
Starch (mg/g DM)	48	36.6

DM : Dry matter

Table 6. Correlations between sugars in pasty grains of sweet grains sorghum

	Glucose	Fructose	Sucrose	Starch
Glucose	1.000			
Fructose	0.996**	1.000		
Sucrose	0.368	0.395	1.000	
Starch	-0.923**	-0.939**	-0.405	1.000

** : Significant correlation at the threshold of 1%

The Pearson correlation matrix (Table 6) reveals very significant correlations at the 1% threshold between the different carbohydrates. Indeed, the glucose content is very significantly correlated positively with the fructose content (r

= 0.996) and negatively with the starch content (r = -0.923). The fructose content was also significantly correlated with the starch content but negatively (r = -0.939). Thus, accessions with high hexose content have low levels of

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Opportunities of preservation of genetic resources for sweet grains sorghum

In the pasty stage of the grains, the panicles are harvested and most part of them is sold to traders. The price of the panicle depends on its size. Thus, the prices charged by these traders generally vary between 0.09 \$ for small panicles to 0.18 \$ for large ones (Figure 3).

Discussion

The large amplitude of variation observed in all the carbohydrates assayed may indicate the existence of genetic diversity within the sweet grains sorghum of Burkina Faso. However, a variation could be genetic or related to a stage of maturation different from the grains of the different harvested accessions (Miguez *et al.*, 2004).

The high content of starch of grains of sweet grains sorghum at the pasty stage compared to the other three carbohydrates (fructose, glucose, sucrose) is similar to the results of previous studies about unsweetened grains sorghum (Ragae *et al.*, 2006; Kulamarva *et al.*, 2009; Kaijage *et al.*, 2014). However, Sweet grains sorghum contains a low sucrose content compared to sweet sorghum, where sucrose is the basis of the sweet taste and predominates with a content of about 80% (Schaffert & Gourley, 1982; Qazi *et al.*, 2012). Sweet grains sorghum is also less rich in glucose than sorghum unsweetened grains, whose content varies from 46.6 to 91.7% (Hanh & Rasper 1973, Nandini *et al.*, 2001). The higher content of fructose compared to sucrose and glucose in grains shows that the sweet taste of grain at this stage in sweet grain sorghum would be due mainly to fructose.

Also, the sweet endosperm character is controlled by a biallelic gene with a dominant effect of an allele (House,

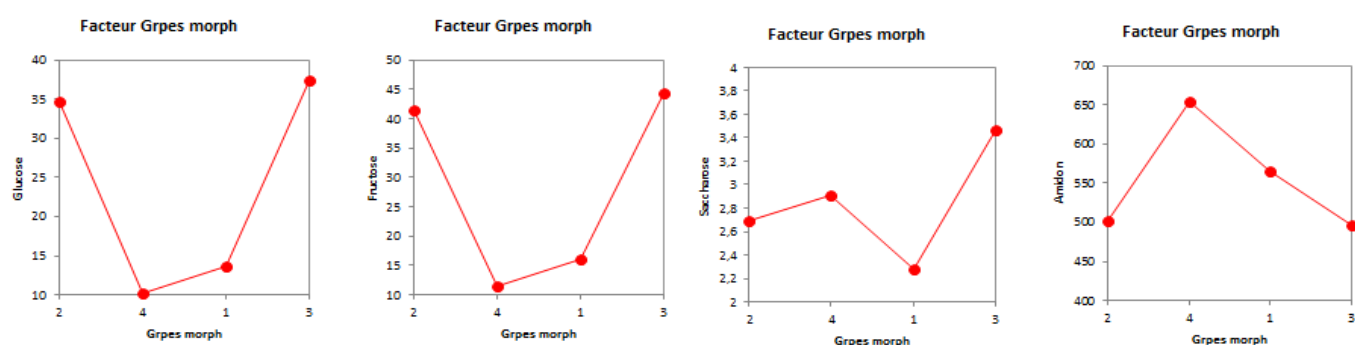


Figure 1. Variation of carbohydrates content in agro-morphological group

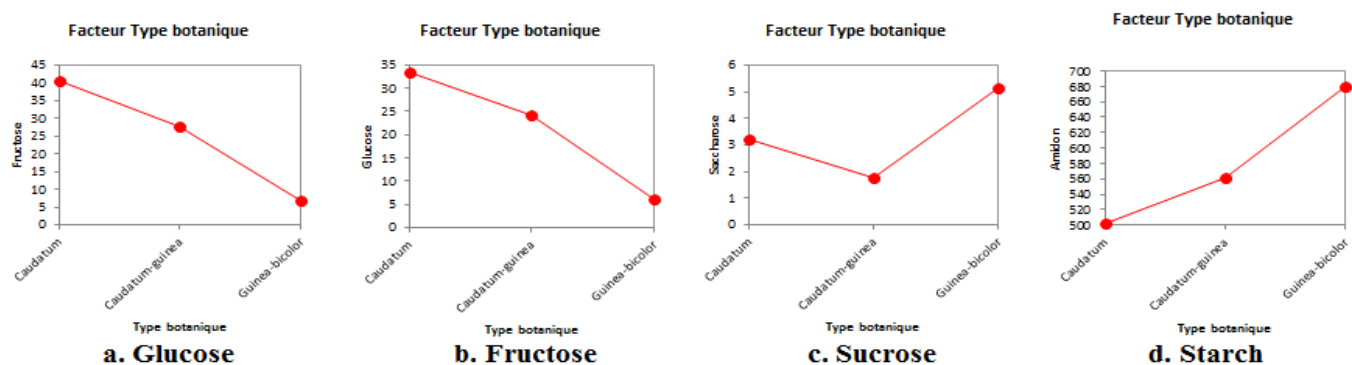


Figure 2. Variation of carbohydrates content in botanical race

1987). The recessive allele "su" codes for a generally high



Figure 3. Sale of sweet grains sorghum panicles

sugar content while the dominant "Su" allele codes for normal sugar content. Thus, the dominant homozygotes "SuSu" and the heterozygotes "Susu" have normal sugar content, while the recessive homozygotes "susu" have an endosperm highly rich in sugar. Sweet grains sorghum would, therefore, be recessive homozygotes of "susu" genotype.

The very significant differences observed between botanical races for glucose, fructose and starch levels also indicate a high variability in the biochemical composition. The *caudatum* race with the highest fructose content and the intermediate race *guinea-bicolor* with the lowest content of fructose would be respectively the sweetest and the least sugared because of the high sweetening power of fructose compared to other carbohydrates.

The variation of the carbohydrate composition according to the agro-morphological groups also testifies to the existence of a genetic diversity. Similar results have already been reported on sweet sorghum (Schaffert & Gourley, 1982; Qazi et al., 2012). Group III would, therefore, contain the sweeter accessions insofar as they are richer in fructose and sucrose. Fructose, sucrose and glucose have sweetening powers of 1.3; 1 and 0.71 respectively.

Finally, the sale of panicles generates monetary income for producers and retail traders. A similar observation had already been reported (Nebié et al., 2012). That increases the purchasing power of producers and resellers and could be contributing to a better preservation of this genetic resource.

CONCLUSIONS

The study revealed that sweetness in the grains of sweet grains sorghum at the pasty stage is due more to fructose than to sucrose. Of the three botanical races, the *caudatum* race is the sweetest while the intermediate race *guinea-bicolor* is the least sweet. It showed also that accessions of the agro-morphological group III are the sweetest and those of group IV are the least sweet.

A better knowledge of the biochemical composition of the grains of this sorghum could make it possible to use them to supplement infant foods in the fight against diseases due to nutritional deficiencies. In addition, the study of the genetic determinism of sweetness by the constitution of a two-parent population and the determination of the mechanism of accumulation of sugars through the biochemical characterization of the whole collection and the kinetics of evolution of the sweet taste during the maturation of the grains could better confirm and deepen the present study and better orient the breeders on the choice of certain genotypes of interest.

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