Degree of Milling Effects on the Sorghum (*Sorghum bicolor*) Flours, Physicochemical Properties and Kinetics of Starch Digestion

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***Abstract*—**Two types of crushing were applied to grains of red sorghum: manual crushing using a mortar and pestle of kitchen and mechanical crushing using a hammer mill. The flours obtained at the end of these various crushing were filtered and subdivided in different fractions according to the diameters of the mesh of the sieves (0.16mm; 0.25mm; 0.315mm; 0.4mm, and 0.63mm…). Some physical, chemical and nutritional traits of these flours were evaluated using Association of Official Analytical Chemists (AOAC). *In vitro* digestibility of these flours was also studied with freezing of flour 1% like substrate and α-amylase from *B. licheniformis* (E.C.3.2.1.1; Megazyme, Wicklow, Ireland). The results revealed that the batches of flours which have the finest diameters as 0.16mm; 0.25mm are the richest one in nutrients and are also the most digestible. Also mechanical crushing is the best mean to obtain significant amount of flours. In conclusion, the type of crushing and the size of the particles have an impact on the final concentration of some nutrients of the flours obtained. Indeed, the finest particles (0.16mm – 0.25mm 0.315mm) obtained after sifting of the flours are more nutritive and have a better digestibility than others size. So the finest particles could be advised for management of cereals namely the sorghum for the production of the infantile foods.

***Keywords*—**Nutrients, digestibility, crush, flour, milling, granulometry.

I. Introduction

IN the field of the food; roots, tubers, and cereals constitute the independent sources of energy brought as starch [1].

Cereals in particular, because of their production's facilities with large scales and due to their digestibility, were considered as better compared to roots and tubers. So more and more, cereals took a capital place in the human and animal consumption [2].

In Asia, rice is the principal produced cereal and constitutes the product of export of many countries (Thailand, China…). In Europe, it is rather the corn which is produced in large quantity with a production of 3.6 million hectares in 2006. It undergoes many transformations and is used in bakery, flour-milling, pastry [3]. As for Africa, it produces mainly grains known as coarse (millet, corn, sorghum…) which are used for the manufacture of many drinks and traditional meals among which one can quote the *dôlo* or the *tchapalo*, the *tôh*, the *dèguê…*[4]. As far as concerned sorghum, it is rich in starch, and the characteristics of its major proteins (kafirins) have been the subject of various studies to understand digestibility properties [5], [6]. Sorghum is also rich in phytochemicals, making it a potential ingredient in the food health, nutraceutical or specialty markets [7], [8]. Currently, sorghum is mainly used for production of weaning foods, where it can be up to 10 % of feed formulations. Particle size reduction is a major pre-process preparation prior to heat-moisture treatments to improve digestibility [9].

The common size reduction machines for grain preparations in food and feed processing include mortar and a pestle of kitchen and modern (hammer, roller and attrition) mills. These mills differ in the effective operating force and the extent of frictional heat generation during grinding [10].

Frictional heat and mechanical energy involved in grinding can affect molecular and structural properties of starch and other components, and influence their functionality [11]-[16]. Some of the measured effects of milling are related to particle size and consequently surface area available for reactions and various end-uses (e.g. amylolysis). Maximization of starch digestion will benefit from knowledge of the mechanisms of digestion to guide processing for an improved utilization of, for example sorghum, in food and feed.

The objectives of this work are to determine which among these two types of milling could be advised for the rural households which use the sorghum as food of weaning and also indicate the type of flour that they should select in the event of sifting of the flours.

II. Materials and Methods

The biological material used consists of grains of red sorghum (*Sorghum bicolor*) which were bought at the market of Port-Bouët, a district of Abidjan Town (Côte d'Ivoire).

A. Preparation of the Samples of Flour

Two kilograms (2kg) bought, are sorted, washed with distilled water and then dried with the drying oven with 45°C during 24 hours until constant weight. A part of the dried

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grains (1kg) was crushed using crushing with hammer of mark TD AFRICA during 35 seconds. It is named crushed red flour of sorghum (CRS).

Another part of the dried grains (1kg) was crushed manually using a mortar and a pestle of kitchen. The rough flour obtained after 30 minutes is called pounded red flour of sorghum (PRS). The flours obtained from these two types of crushing are stored in plastic pots to be granulated (electric sifting).

A sample of 800g flour is deposited at the top of a stacking of 16 sieve whose mesh size goes decreasing (5mm – 3.15mm - 2mm – 1.25mm – 0.8mm – 0.63mm – 0.5mm – 0.4mm – 0.315mm – 0.25mm – 0.2mm – 0.16mm – 0.125mm – 0.1mm – 0.063mm – 0.01mm) and to which one applies a mechanical vibratory movement during 10 minutes. The particles whose dimensions are lower than the one of the openings cross the sieve when it is put in vibration (the passerby), meanwhile the largest particles are retained (the refusal). The particles are distributed in an unequal way on each sieve. The fractions retained on each sieve are then weighed to determine the granulometric distribution.

This operation of electric sifting is carried out distinctively for the flour of crushed red sorghum and that of pounded red sorghum.

The fractions of flours lately obtained according to their size are named (Fig. 1):

"Pounded Red flour of Sorghum composed of particles of 0.16mm of diameter: $PRS_{0.16}$"

"Crushed Red flour of Sorghum composed of particles of 0.16mm of diameter: $CRS_{0.16}$"

"Pounded Red flour of Sorghum composed of particles of 0.315mm of diameter: $PRS_{0.315}$"

"Crushed Red flour of Sorghum composed of particles of 0.315mm of diameter: $CRS_{0.315}$"

They are then preserved in various plastic bottles for later analyzes.

B. Extraction of Ethano-Soluble Sugars

Soluble sugars were extracted from a sample of malt flour mixed with ethanol solution (80% v/v) that had been agitated for 30min in a thermostated bath at 90°C and then centrifuged at 5000rpm for 10min at 4°C. The supernatant was retrieved and the same procedure applied to the residue. The two mixed supernatants were dry evaporated overnight using a Speedvac centrifugal evaporator (JOUAN RC 10.10, Saint Herblain, France), then stored at 4°C, before the determination of sugar contents.

C. Evolution of Physicochemical Components

The content of dry matter and ash was determined by the method of the AOAC [17]. The content of soluble sugar was determined by method of Dubois et al., [18]. Reducing sugar was estimated according the method of Bernfeld [19] using glucose as standard. The total glucids are proportioned according to Bertrand and Thomas [20]. Total starch content was measured by AACC method 76.13 B [21].

D. Digestibility of Starch

For studies relating to starch digestion, α-amylase from *B. licheniformis* (E.C.3.2.1.1; Megazyme, Wicklow, Ireland) supplied at a concentration of $3000 UmL^{-1}$ was added to the gel of sorghum flour containing. Glucoamylase from *Aspergillus niger* (E.C. 3.2.1.3; A7095, Sigma, St Louis, MO) was obtained at a concentration of $300 UmL^{-1}$, where a single unit of enzyme is defined as that amount which hydrolyzes the α (1,4) linkage of maltose at a rate of $1 mmolmin^{-1}$, at 25°C. After appropriate dilution of either one of the enzymes was added to a flour suspension. The rates of hydrolysis of starch were measured. Both were pretreated with a cocktail of hydrolytic enzymes [22] including porcine pancreas α-amylase (A4268, Sigma), porcine mucosa pepsin (P7000, Sigma), porcine pancreas pancreatin (P7545, Sigma) and glucoamylase. The mixture was incubated with stirring in a water bath at 37°C for 100min. The glucose released as a result of starch digestion was measured with an AccuCheck® Performa® glucometer (Roche Diagnostics Australia Pty. Ltd., Caste Hill NSW 2154, Australia), and digested starch (g per 100g dry starch) at a measurement time (min) was calculated as before [22].

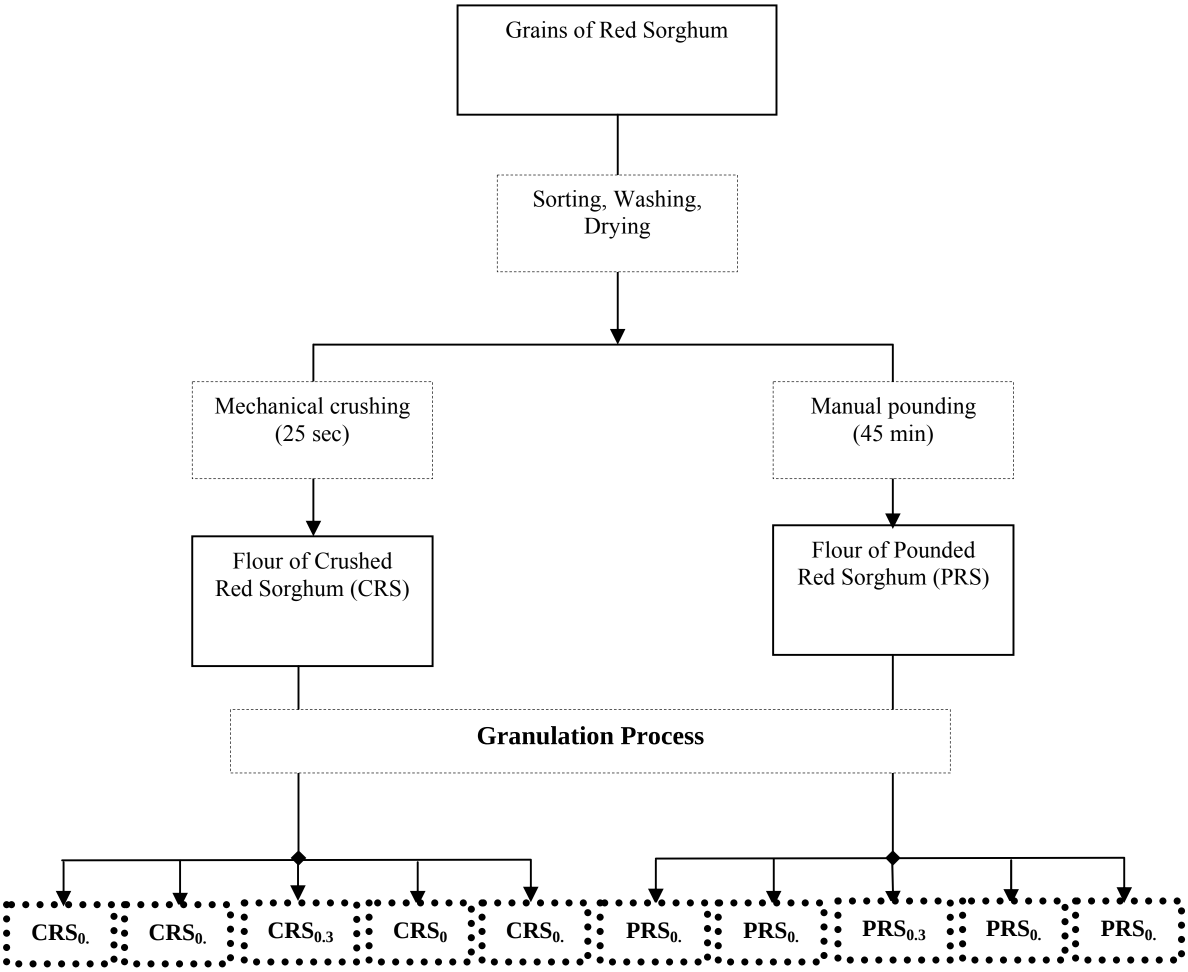

Fig. 1 Diagram of flours production

E. Statistical Analyses

The statistical analysis of the results was made by the software Stat Soft (statistica, 99ème Edition France). The significance of the differences between the various samples of flours was calculated with the test of DUNCAN on the level of significance 0.05.

III. Results and Discussion

A. Granulometric Distribution of the Flours after Crushing

Fig. 2 shows the granulometric distribution of the flours pounded from the red sorghum and the flour crushed from red sorghum. The flour pounded from red sorghum is primarily made up of 84.98% of particles of sizes ranging between 1.25 mm and 0.63mm diameter. The particles of sizes ranging between 0.5mm and 0.06mm diameters are slightly represented (13.25%). As for the flour of crushed sorghum by the machine, it is made up in majority of particles of sizes comprised between 0.8mm and 0.5mm diameter with a percentage of 65.01%. The particles of size ranging between 0.5mm and 0.06mm diameters account for 26.91% there.

The analysis of the granulometric distribution of the flours shows that the red flour of sorghum crushed with machine (CRS) is finer than that resulted from pounded sorghum (PRS). These results can be explained by the difference in effectiveness between both methods of crushing. Indeed, hammer crushing acts at the same time by impact (action of the hammers) and by abrasion (action of the grids). The finest particles result into major part of abrasion and the coarsest particles of the impact.

The proportion of grains crushed by abrasion is most important.

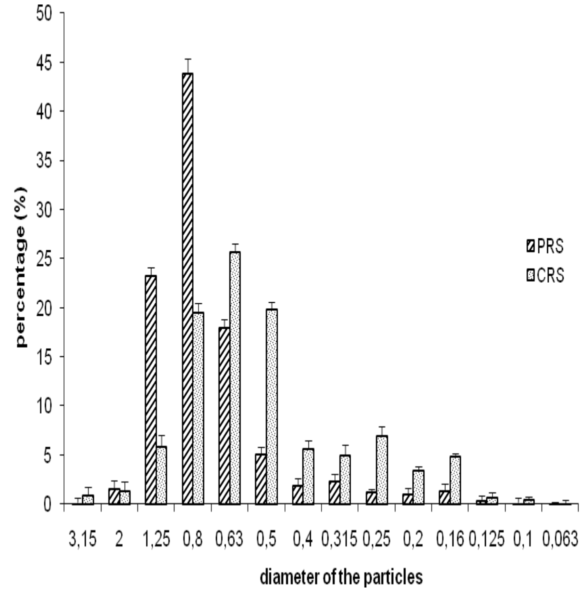

Fig. 2 Granulometric distribution of the flours after crushing PRS: Pounded Red flour of Sorghum; CRS: Crushed Red flour of Sorghum

B. Chemical Composition of the Flours of Red Sorghum after Crushing

Fig. 3 presents the concentration in some nutrients of the flours obtained after mechanical crushing (CRS) and manual crushing (PRS). The contents of dry matter (DM), total glucids (TG) and starch are very important (≥75%). Moreover, concentrations of total sugar (TS) reducing sugar (RS) and the mineral matter (MM) are very weak (≤5%) no matter the type of crushing is. The flour obtained after manual crushing contains high out of dry matter, reducing sugars and total sugars compared to that obtained after mechanical crushing. On the other hand, the contents of total minerals, glucids and starch are more important in the flour obtained after mechanical crushing. The type of crushing has then an effect on the chemical composition of the red sorghum. Moreover, inside the hammer mill, the forces of pressure applied to a matter grain create a three-dimensional field of constraint set out again in a continued way in the volume of material. Yet, in the case of crushing with the mortar, the shocks of the pestle on the grains cause a stress field set out again in a uniform way on the vertical. What provokes less bursting of the grains crushed with the mortar to the grains crushed with the machine [23].

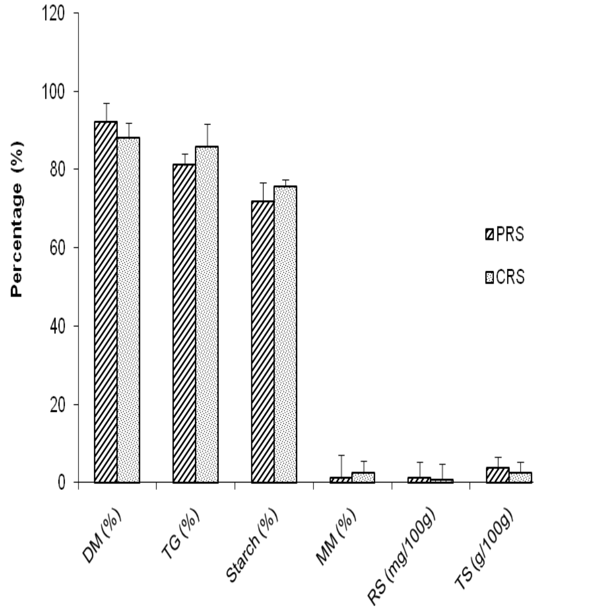

Fig. 3 Effect of the type of crushing on some nutrients of the red flour of sorghum DM: Dry Matter; TG: Total Glucids; MM: Total Ash; RS: Reducing Sugar; TS: Total Sugar PRS: Pounded Red flour of Sorghum; CRS: Crushed Red flour of Sorghum

C. Effect of the Size of the Particles on the Chemical Composition of the Flours

Table I showed chemical composition of each particle size. The results reported here showed that the separation of the flours in homogeneous particles (granulation or sifting) has a variable effect on the concentration in it nutrients. Thus, we observe a reduction in the percentages out of dry matter, reducing sugars, total sugars followed by an increase in the percentages in total ashes, total glucids and starch of the red flour of sorghum crushed, contrary to the percentages of dry matter, reducing sugars which undergo an increase and a reduction in the percentages out of reducing sugars, total glucids and starch of the red flour of sorghum crushed. Whatever the type of crushing is, the finest flours (0.16mm – 0.25mm – 0.315mm) present higher concentrations of the percentages out of dry matter, mineral matter, reducing sugars, total glucids and out of starch. The values lie between 87% - 89% of dry matter, 3% - 5% of ashes, 0.82% - 1.62% of reducing sugars, 83% - 89% of total glucids, and 72% - 74.18% of starch. However, the coarsest flours (0.4mm and 0.63mm) contain weaker concentrations in these quoted known nutrients. The values lie between 86% - 87% of dry matter, 1% - 2% of ashes, 0.23% - 0.8% of reducing sugars, 80% - 82% of total glucids and 70.21% - 71.27% of starch. As for the coarsest flours, they are much richer in total sugars. The values lie between 1.68% - 2.7% of total sugars in the flours of 0.4mm and 0.63mm of diameter and 0.6% - 1.77% in the flours of 0.16mm; 0.25mm and 0.315mm diameter. These results are in agreement with those of Carré [24]. The higher dry matter concentrations, reducing sugars and total sugars in the flour of PRS and the more important concentration out of mineral matter in the flour of CRS are explained by the modification of the constitution interior of the particles under the action of the mechanical constraints and the temperature. The strong concentration out of dry matter of the flour of PRS is due to the phenomenon of dehydration which is more important at the time of the process of pounding. These results are confirmed by Reference [25] who showed that pounding process causes a more important water loss by drying of the flours because of duration of the operation and the rise in the temperature.

The high values of the concentration out of RS and TS of the flour of PRS are explained according to Reference [26] by the disorganization of the structure of the starch contained in the cellular grains and walls during the pounding. These authors showed that, during crushing, the disturbance of fabrics of the cellular walls increases the sugar rate. The raised value of ashes in the flour of CRS is explained by the effect of the temperature on the concentration in ashes. The mechanical crushing being of short duration, the rise in the temperature of the flour of CRS is less long-term. Certain ashes being volatile in contact with heat, the mineral concentration will be consequently higher in the flour of CRS than in that of PRS [27].

TABLE I
INFLUENCE OF SIZE OF PARTICLES ON THE CONTENTS OF SOME NUTRIENTS OF THE RED SORGHUM FLOUR

Samples	Dry matter (%)	Total Ashes (%)	Reducing Sugars (%)	Total Sugars (%)	Total Glucids (%)	Starch (%)
PRS	92.00 ± 1.12^{d}	1.10 ± 0.36^{a}	1.22 ± 0.10^{ef}	3.63 ± 0.15^{g}	81.46 ± 1.3^{ab}	71.31 ± 10^{abc}
CRS	88.00 ± 11^{abc}	2.51 ± 0.42^{cd}	2.36 ± 0.15^{h}	0.71 ± 0.09^{a}	85.75 ± 0.7^{cd}	75.30 ± 1.10^{e}
$PRS_{0,16}$	92.00 ± 1.12^{d}	3.03 ± 0.15^{d}	0.53 ± 0.20^{b}	1.40 ± 0.20^{c}	84.34 ± 0.80^{cd}	71.31 ± 10^{abc}
$CRS_{0,16}$	89.33 ± 0.60^{c}	3.04 ± 0.41^{d}	1.03 ± 0.15^{de}	1.05 ± 0.05^{b}	84.11 ± 1.00^{cd}	74.15 ± 0.90^{de}
$PRS_{0,25}$	88.00 ± 1.01^{abc}	5.05 ± 0.32^{e}	0.82 ± 0.02^{cd}	1.00 ± 0.10^{b}	87.43 ± 2.30^{ef}	72.32 ± 1.05^{abcd}
$CRS_{0,25}$	89.00 ± 1.30^{bc}	5.01 ± 0.52^{e}	1.25 ± 0.25^{efg}	1.77 ± 0.03^{d}	89.19 ± 0.90^{f}	72.11 ± 2.02^{abcd}
$PRS_{0,315}$	87.00 ± 1.31^{ab}	2.56 ± 0.40^{cd}	1.03 ± 0.06^{de}	0.60 ± 0.10^{a}	83.01 ± 0.10^{bc}	73.14 ± 1.00^{bcde}
$CRS_{0,315}$	$89,33\pm 0.60^{c}$	1.51 ± 0.42^{a}	1.63 ± 0.13^{g}	2.70 ± 0.20^{f}	80.61 ± 0.51^{a}	72.23 ± 1.01^{abcd}
$PRS_{0,4}$	$86,66\pm0.50^{a}$	2.11 ± 0.31^{bc}	0.64 ± 0.14^{bc}	1.68 ± 0.16^{d}	80.00 ± 11.05^{a}	72.00 ± 2.02^{abcd}
$CRS_{0,4}$	87.00 ± 20^{ab}	2.11 ± 0.52^{bc}	0.23 ± 0.05^{a}	2.14 ± 0.04^{e}	80.12 ± 0.93^{a}	71.27 ± 0.91^{ab}
$PRS_{0,63}$	86.33 ± 0.50^{a}	1.02 ± 0.21^{a}	1.44 ± 0.34^{fg}	1.76 ± 0.15^{d}	82.57 ± 0.55^{bc}	70.61 ± 0.50^{a}
$CRS_{0,63}$	86.00 ± 1.90^{a}	1.10 ± 0.11^{ab}	0.81 ± 0.11^{cd}	2.71 ± 0.22^{f}	83.07 ± 11.01^{bc}	70.21 ± 1.01^{a}

These indicated values in the table are the average of three measurements. For a given nutrient, the same letter registered in the same column indicates that the values present no significant difference between the samples with the threshold 5%. PRS: Pounded Red flour of Sorghum; CRS: Crushed Red flour of Sorghum. Values are means (± SEM). Means not sharing a common superscript letter in column are significantly different at ($p \leq 0.05$) according to Duncan's multiple range tests.

D.Effect of the Type of Crushing and Granulometric on in vitro Digestibility of Flour of Red Sorghum

Fig. 4 shows digestibility data for size fractions of milled sorghum grains, given as a fraction of total starch digested after 100min of incubation. No matter what the type of crushing, the percentage of digestibility increases with time. It ranges between 1.06% and 60% at the most. The filtered flours (flours made up of homogeneous particles) have a better digestibility than those of the not filtered flours (flours contained homogeneous particles). The percentages of digestibility lie between 1.06% - 57.76% and 2.59% - 17.86% respectively for the filtered flours and the not filtered flours. These results indicate that the type of crushing has a direct impact on the digestibility of the flours. Indeed, no matter what the size of the particles is, the flours crushed with the machine are much more digestible than those pounded with pestle. The values are in particular included between 10.43% - 29.9% and 1.06% - 8.67% respectively for the flour of $CRS_{0,315}$ and the flour of $PRS_{0,315}$. Also, the smaller the particle size, the more digested was sorghum, presumably due to an increase in relative surface area [10]. Digestion of starch is dependent on starch properties such as granule size, architecture, crystalline pattern, degree of crystallinity, surface pores or channels, degree of polymerization, others components except the starch and their interactions with starch, and amylose/amylopectin ratio [28]-[32]. Although reduction of particle size would be the main effect of grinding, depending on the degree of grinding, ground starch could lose its crystallinity and yield low molecular weight materials [12], [16]. Also, hammer mills and mortar differ in frictional heat during grinding, and this could be an additional factor that affected starch digestion in the samples. The values of digestibility start from 12.28% - 35.17% and 3.36% - 26.5% respectively for the flours of $CRS_{0,63}$ and the flour of $PRS_{0,63}$ to 13.65% - 57.76% and 1.21% - 32.20% respectively for the flour of $CRS_{0,16}$ and the flour of $PRS_{0,16}$.

The study of *in vitro* digestibility of the flours of red sorghum initially showed that it ranges between 1% and 60%. This value is very average compared to the digestibility of simple sugars (glucose, saccharose) which is close to 100% [2]. That is explained by the presence of a significant amount of indigestible carbohydrates (fibers), and polyphenols compounds (tannin) in the flour of sorghum. What contributes to the reduction of the digestibility of the flour of sorghum [25]. Then, the results showed that the filtered flours are more digestible than the one non filtered [33]. That can be explained by the fact why the simultaneous presence of the very fine and coarse particles could have an antagonistic effect on gastric motricity [24]. As, the results revealed as for identical sizes of particles, the digestibility of the flour of CRS is significantly more important than that of PRS. That could be explained by the difference in hardness of the particles after crushing. Indeed Reference [34] showed that a food particle can be defined by its size, its form, or its color, its density, its elasticity, its roughness and especially its hardness; and that digestibility the particle is related to these parameters. The flours having undergone various types of crushing, they have different hardness, with a more important hardness for the particles of PRS compared to the particles of CRS because of effectiveness of the operations. Reference [35] as showed as for two identical pea flours of granulometry being different by mechanical energy necessary to their crushing, it is resulted from it, without reduction in granulometric size, a very clear improvement of the digestion of the starch with the flours having required a higher mechanical energy. So far, the results showed that digestibility increases linearly with the reduction of the size of the particles. That is confirmed by the work of Reference [36] and those of Reference [35] which revealed that the granulometric size after crushing is positively correlated with hardness. In other words, after crushing, the finest particles are the least hard. They are consequently more digestible. Also, according to Reference [37], due to their less hardless, the finest particles diffuse more easily in the soluble fraction of the food matter and are consequently more easily digested. In the same way, References [2] and [38] showed that the CUD of the starch decreases when the content amylose but also the size of the grain increases.

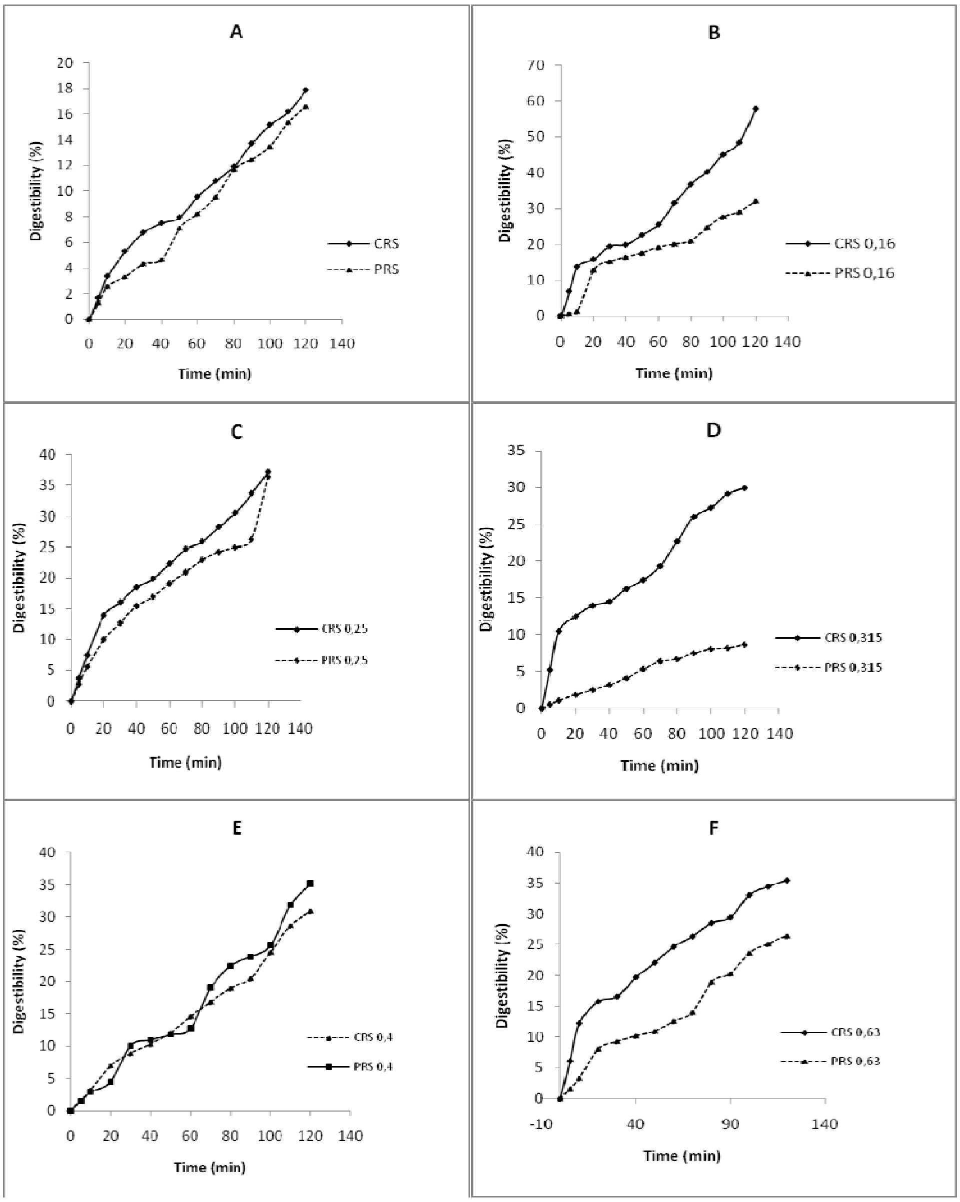

Fig. 4 Evolution of the starch's *in vitro* digestibility in the flours resulted from unfiltered pounded red sorghum (PRS) and crushed red sorghum (CRS). (A): PRS: Pounded Red flour of Sorghum; CRS: Crushed Red Flour of Sorghum; (B): Flours particles with diameter of 0.16mm: (C) Flours particles with diameter of 0.25mm; (D) Flours particles with diameter of 0.315; (E) Flours particles with diameter of 0.4mm and (F) Flours particles with diameter of 0.63mm

IV. Conclusion

Under the terms of our study, we can say that the type of crushing and the size of the particles have an effect on the final concentration of certain nutrients of the flours obtained. Mechanical crushing has two major advantages: it is less hard and the flour resulting from this type of crushing is more digestible than others. Also, the finest particles (0.16mm – 0.25mm 0.315mm) obtained after sifting of the flours are more nutritive and have a better digestibility. They are obtained in greater number by mechanical crushing. Because of the quoted known advantages, mechanical crushing followed of a sifting to recover the finest particles could thus be advised with manage which use cereals in particular the sorghum to produce the infantile flours.

Acknowledgments

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