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Effect of milling and sieving processes on the physicochemical properties and flow properties of okra powders

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Keywords:	okra seeds, sieving, particle size, flowability, FT4 powder rhometer, Milling

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Graphical abstract

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Effect of milling and sieving processes on the physicochemical properties and flow properties of okra powders.

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Abstract

The aim of this work was on one hand to investigate the influence of milling speed and mean particle size of granulometric classes of okra (*Abelmoschus esculentus* (L.) Moench) seed powders on proximal composition (related to nutritional potential). On the other hand, it was tried to relate flow properties to powder physicochemical characteristics, namely particle size distribution and proximal composition. Then, dry okra seeds were ground with an ultra-centrifugal mill at three different rotational speeds: 6 000, 12 000, and 18 000 rpm. Each powder was separated into four granulometric classes: $\geq 500 \mu\text{m}$, 315 - 500 μm , 180 - 315 μm , and $\leq 180 \mu\text{m}$. Milling speed affected the particle size distribution of dry okra seed powders: on one hand, the higher the milling speed, the lower the mean particle size, mainly because of the higher the proportion of fine particles ($< 100 \mu\text{m}$); on the other hand, at low milling speed, dry okra seed powders were rather monomodal and constituted of large particles, whereas at high milling speed they were bimodal and composed of two populations (fine and large particles). In granulometric classes, fat and protein contents were significantly higher for fine particles, unlike carbohydrate and ash contents that were lower. This differential distribution of macronutrients within granulometric classes was enhanced at higher milling speed. Finally, the $\leq 180 \mu\text{m}$ granulometric class, corresponding to the smallest mean particle size, had a low flowability, confirmed by their high cohesion and compressibility, which can be related to its high fat content making it sticky. On the contrary, the $\geq 500 \mu\text{m}$ granulometric class was not cohesive, leading to good flow properties.

Key words: okra seeds; milling; sieving; particle size; FT4 powder rheometer; flowability

1. Introduction

Okra (*Abelmoschus Esculentus* (L.) Moench) is a vegetable widely grown in tropical and subtropical regions (Torkpo *et al.*, 2009; Jarret *et al.*, 2011). It is called bamia or bamya in some parts of the world, particularly in Asia and the Middle East (Uka *et al.*, 2013). This crop is generally considered as drought-resistant and has fewer pests than soybean (Miller *et al.*, 1949; Jarret *et al.*, 2011). Okra plays an important role in the human diet by supplying carbohydrates, protein, fat, minerals, and vitamins that are usually deficient in staple foods (Adelakun *et al.*, 2009). It is a vegetable with an enormous economic potential for poor communities because of its sales potential in rural and urban markets (Adelakun *et al.*, 2011). Although the seed pods are most often used (Camciuc *et al.*, 1998; Ndangui *et al.*, 2010), mature seeds are known to have superior nutritional quality. (Oyelade *et al.*, 2003) reported that okra seed is rich in proteins and lipids. A Nigerian research program initiated by Nigeria reported that dry okra seeds were a good source of proteins (45 %) and fat (34 %) (Oyelade *et al.*, 2003; Adelakun *et al.*, 2009), classifying it as a nutrient-rich plant, which could be used for feeding children and fight against malnutrition. The high nutritional potential of okra seeds was also evidenced. In the Republic of the Congo, research about okra seeds began in 2010 with Ndangui and Kimbonguila Ndangui *et al.* (2010), who determined the proximate composition of okra seeds (24 % proteins, 24 % fat, 36 % carbohydrates), and were concluded that okra have high nutritional potential. The nutritional importance of okra seeds reawakened interest in bringing the crop into commercial production. Although extensive information is available about nutritional quality of okra seeds, no study, to our knowledge, focused on the distribution of nutritional compounds within granulometric classes of okra seed powders produced by milling processes. In fact, it can be hypothesized that large particles may be richer in fibers, which are difficult to grind; hence, small particles may be enriched in minerals, fat, and proteins and have a higher nutritional quality, as previously reported for various plant parts (Becker *et al.*, 2016; Zaiter *et al.*, 2016; Deli *et al.*, 2019a). For this purpose, the current study deals with the production of okra seed powders by successive milling and sieving processes, with the aim to determine if some granulometric classes of okra seeds powder could both have a great nutritional quality and good flow properties (Adelakun *et al.*, 2011; Chen *et al.*, 2015), making them suitable for the production of food supplements. Milling induces particle size reduction, broadening of particle size distribution, and increases specific surface area, leading to the improvement of material functionalities, especially regarding extraction efficiency and bioavailability of nutritional bioactive compounds (Crayveld *et al.*, 2009; Zhao *et al.*, 2009; Zhang *et al.*, 2012; Zaiter *et al.*,

2016). The combination of milling and sieving processes could be interesting to identify granulometric classes richer in fat and proteins, ~~generally concentrating bioactive compounds~~ (Brewer *et al.*, 2014; Becker *et al.*, 2016; Lucas-González *et al.*, 2017). ~~This would enable optimizing the extraction of bioactive compounds of plants, by reducing solvent consumption and extraction time (Zaiter *et al.*, 2016; Deli *et al.*, 2019a).~~

~~Particle size control seems also interesting with regard to powder flowability~~ Nutritional quality of food powders is not the only criterion for their production at industrial scale, their ability to be transported, handled, and stored is also crucial for their manufacturing. Powder flowability is one of the most important powder technofunctionalities regarding these aspects. Thus, this study intended to evaluate the nutritional quality but also the flowability of granulometric classes of okra seed powders. It has been shown that particle size distribution is a major characteristic controlling the flowability of powders having similar proximate composition (Petit *et al.*, 2017). A decrease in particle size is often associated with an increase in cohesion because the interparticular forces are increased when the distance between particles is reduced owing to higher interparticular surface contact area. So, small particles are generally cohesive and have a poor flowability ~~lower capacity to flow~~. The major influence of particle size on flow behavior has been recently reported for many powdered foods and food supplements: for instance, *foufou* and *foutou* powders (instant plantain-based powders) (Gnagne *et al.*, 2017), *Boscia senegalensis*, *D. glomerata* and *H. sabdariffa* plant powders (Deli *et al.*, 2019b).

Thus, the purpose of the present study was to investigate the nutritional quality on the basis of proximal composition and flow properties (flow energy, compressibility, cohesion, flow factor, etc.) of these powders, as powder flowability is an important functionality for industrial manufacturing, handling, and storage. This was achieved with the FT4 powder rheometer (Freeman Technology) (Gnagne *et al.*, 2017), which provides comprehensive series of methods to characterize powder behavior. In this study, okra seeds were milled with an ultra-centrifugal mill at three rotating speeds: 6 000, 12 000, and 18 000 rpm, then the three powder samples were each separated in four granulometric sizes by sieving: $\geq 500 \mu\text{m}$, 315 - 500 μm , 180 - 315 μm , and $\leq 180 \mu\text{m}$. The influence of proximal composition and mean particle size of granulometric classes of okra seed powders on their flowability was investigated, in order to conclude on the impact of successive milling and sieving processes on okra seed powder nutritional quality and flow characteristics.

2. Material and methods

2.1. Material: okra seeds

Okra seeds were purchased from farmers in Impfondo locality, in the north of the Republic of the Congo (1°37'7.47"N; 18°3'44.459"E). Okra were shelled (the seeds were removed from the pods), then seeds were sorted to remove impurities and sun-dried in the open air at 30 °C. Dry seeds were stored at 10 °C until powder production.

2.2. Powder production by milling and sieving

500 g sun-dried okra seeds were milled with an ultra-centrifugal mill ZM 200 (Retsch, France) at three different milling speeds (6 000, 12 000, and 18 000 rpm) at room temperature (25 °C) using a 20-cm sieve drilled with 1-mm trapezoidal holes. Then, obtained powder was collected in sealable polyethylene bags before performing sieving.

2.3. Okra seed powder sieving

Okra seed powders issued resulting from the milling step were sieved with an Analyzer 3 Spartan sieve shaker (Fritsch, Idar-Oberstein, Germany) with 0.5 mm vibration amplitude for 24 min using sieves of 180, 315, and 500 µm mesh sizes. Hence, the following granulometric classes were produced: ≥ 500 µm, 315 - 500 µm, 180 - 315 µm, and ≤ 180 µm.

The material on each sieve was collected in sealable polyethylene bags, weighed to determine the mass yield of each granulometric class, and stored at 10 °C until analysis.

2.4. Physical properties of okra seed powders: particle size distribution

The particle size distribution of okra seed powders (unsieved powders and granulometric classes) were determined by laser diffraction granulometry using dry dispersion (Mastersizer 3000, Malvern Instruments France, Orsay, France) at room temperature (25 °C). Dispersion conditions for all powders except the ≤ 180 µm powder samples were as follows: 100 % air pressure at 4 bar, 3 mm hopper gap, and 40 % feed rate. For the ≤ 180 µm powder samples, the following dispersion conditions were employed: 100 % air pressure at 4 bar, 4 mm hopper gap, and 70 % feed rate. The chosen size estimator was the equivalent diameter in volume.

The d10, d50, and d90, which are characteristic diameters representative of the particle size distribution, were calculated. They represent the diameters for which 10 %, 50 % and 90 %, respectively, of the particle population have lower sizes. Last, the span was used to evaluate the width of the particle size distribution; it was determined according to the following equation 1:

$$\text{Span} = \frac{d_{90} - d_{10}}{d_{50}} \quad (\text{Eq. 1})$$

2.5. Powder proximate composition

The proximate composition of all granulometric classes as well as unsieved powders were evaluated with standard methods. Fat content was determined by Soxhlet method using 5 g

powder, hexane as extraction solvent, and 8 h extraction duration (Castro *et al.*, 1998). Moisture content was measured by weight loss after drying 3 g powder at 103 °C for 5 h (AFNOR (Agence Française de NORmalisation), 1976). Ash content was evaluated by incinerating 3 g powder in a muffle furnace at 550 °C for 12 h (ADPI (American Dry Product Institute, Elmhurst, IL), 2002). Total protein content was determined by Kjeldahl method (Bradstreet, 1954). Carbohydrate content was calculated as the difference between 100 % and the contents in other components; the associated standard error was deduced by propagation of uncertainties.

2.6. Characterization of powder flow properties

Rheological properties of okra seed powders were measured with the FT4 powder rheometer (Freeman Technology, Worcestershire, UK) using 25 mm accessories. Measurement test of stability, compressibility, aeration, and shear cell were performed with the standard method of powder rheometer FT4 with some modifications (Gnagne *et al.*, 2017). Detailed descriptions of FT4 standard tests can be found in Freeman (2007) and Leturia *et al.* (2014).

2.6.1. Stability test

The stability test is intended to determine if powder flowability in unconfined (low normal stress) conditions is affected when making the powder flow. Indeed, powder agglomeration, segregation, or attrition may change the energy needed to make the powder flow. Thus, the energy required to make the powder flow was measured in flow conditions imposed by the standard stability test seven times successively by rotating a blade down and up in the conditioned powder bed. Thus, the stability test consists in the succession of test and conditioning cycles. Conditioning was achieved by the gentle displacement of the powder bed by the rotating blade. Conditioning and test cycles were carried out at 100 mm/s blade tip speed and the powder sample was loaded into an assembly of two glass cylinders, the powder bed was conditioned and then the upper cylinder was removed to obtain a precise volume (25 mL) of conditioned powder for tests. From the measurements of force and torque of the blade, various flow parameters can be extracted; the most important were:

- Basic flowability energy (BFE, mJ), defined as the flow energy measured upon downward moving of the blade during test 7.
- Specific energy (SE, mJ/g): it defines the energy per gram needed to displace the conditioned powder bed during upward moving of the blade. It represents a measure of how powder flows in a low-stress environment.

- Conditioned bulk density (CBD, g/mL), which is a density measurement of the powder bed in conditioned state. It is thus very similar to the classical powder aerated density.

2.6.2. Compressibility test

For the compressibility test, the powder sample was first placed in the measurement cell (i.e. two overlaid 25 mm × 10 mL glass cylinders) and submitted to three conditioning cycles. After that, the vessel was split to remove any excess powder and the air was replaced by the vented piston. Then, powder was slowly compressed by the vented piston moving vertically at 0.05 mm. s⁻¹ at different normal stresses ranging from 0.5 to 15 kPa, and the percentage change in volume of the powder bed, corresponding to powder compressibility, was measured. Powder compressibility at 15 kPa CP_{15kPa} was used to calculate compacted density (CD_{15kPa}), which is the mass of powder per unit volume of the powder bed, including the volume of all interparticle voids (Abdullah and Geldart, 1999) when applying 15 kPa normal stress. CD_{15kPa} was calculated with equation 2:

$$CD_{15kPa} = \frac{100}{(100 - CP_{15kPa})} \cdot CBD$$

2.6.3. Aeration test

The aeration test is a quick way to evaluate the fluidization ability of a powder. The aeration test consists in introducing air into the powder and evaluating how this changes the flow energy (Freeman, 2007; Leturia *et al.*, 2014; Gnagne *et al.*, 2017). The powder sample was placed in the measurement cell (25 mm x 35 mL glass cylinder) fitted with a porous plate and three conditioning cycles were carried out to achieve a homogeneous packing state of the powder sample. The aeration test was then performed by measuring the flow energy of the powder bed during downward moving of the blade through the powder bed at 100 mm.s⁻¹ tip speed for air velocities ranging from 0 to 5 mm s⁻¹ by 0.5 mm. s⁻¹ steps. Between each test cycle, a conditioning cycle was carried out to obtain a steady state of aerated powder. The most important parameters measured by the aeration test are defined below:

- Aerated energy (AE, mJ), which is the flow energy measured during downward moving of the blade through the powder bed at maximal air velocity (10 mm.s⁻¹).

- Aeration ratio (AR, dimensionless) that designates the factor by which the basic flowability energy (i.e. at 0 mm.s⁻¹ air velocity) is reduced when aerating the powder bed at maximal air velocity of 10 mm s⁻¹. It was calculated according to equation 3:

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$$AR = \frac{BFE}{AE} \quad (\text{Eq. 3})$$

2.6.4. Shear cell test

Shear cell tests are intended to determine the shear stress required to initiate flow in a pre-consolidated powder, using a specially designed shear head equipment. This test provides indication of how easily a powder moves from a static state to dynamic flow in confined environments (i.e. at high normal stress). During the FT4 shear cell test, the sample powder is subjected to a pre-consolidation step at 9 kPa normal stress before subsequent shear tests are carried out at normal stresses ranging from 7 to 3 kPa by 1 kPa steps (Freeman, 2007; Yu *et al.*, 2011; Fu *et al.*, 2012), in which the shear stress required to cause failure of the pre-consolidated powder bed (thus making the powder flow) is measured. This succession of shear tests at decreasing applied normal stresses permits to build the yield locus of the normal stress vs. shear stress curve, from which powder flow properties such as flow factor and powder cohesion can be derived.

Cohesion, expressed in kPa, represents the shear stress required to make the powder bed flow in unconfined conditions: indeed, the cohesive interactions between particles must be broken before they can be individually put in motion. Moreover, generally, the higher the interparticular forces, the more the powder bed can be compressed, and the more energy must be supplied to the powder to make it flow.

Flow factor (ffc) is defined as the ratio between the consolidation stress and unconfined yield stress: The larger the ffc value, the better the powder flows (Jenike, 1964; Rhodes, 2008). Powder flow behaviors can be classified as follows (Fu *et al.*, 2012):

ffc < 1: not flowing;

1 < ffc < 2: very cohesive;

2 < ffc < 4: cohesive;

4 < ffc < 10: easy flowing;

10 < ffc: free flowing.

2.7. Statistical analysis

All experiments were conducted in triplicate and results were expressed as mean $\pm$ standard deviation. Statistical analysis was performed to a one-factor analysis of variance using Microsoft Excel 2016, significant differences were assessed with Tukey's HSD at a 5 % confidence level with the DSAASTAT add-on.

3. Results and discussion

3.1. Impact of milling speed on okra seed powder granulometry

Particle size distributions of okra seed powders obtained at the three milling speeds (6 000, 12 000, and 18 000 rpm) are shown in figure 1.

Figure 1

First, it was observed that okra seed powders were constituted of two populations: one of large particles between 200 and 3 000 μm , the other of fine particles between 5 and 200 μm . This is typical for seed powders obtained by milling, as the different organs of seeds (seed coat, radicle, cotyledon, etc.) differ in hardness, thus resulting in various particle sizes (Chen *et al.*, 2015). Then, the shift to the left of the particle size distribution at higher milling speed shows that it led to smaller particles, as expected. Besides, when the milling speed was increased, the proportion of fine particles raised to the detriment of the proportion of large particles. This shows that some large particles, presumably having elevated hardness, were converted in finer ones at higher milling speed. Also, it can be guessed that the employed ultra-centrifugal mill may not allow decreasing the particle size under 10 μm , in agreement with equipment specifications. Moreover, figure 1 confirms the effect of the milling speed on the particle size distribution of okra seed powders. It is worth noting that a more intense milling was also associated to a broader dispersion of the particle size distribution. Indeed, obtained powder was less monomodal when milling speed was increased, as evidenced by the increase in span. This may be indicative of the occurrence of different grinding mechanisms in the employed milling process. From this granulometric analysis, three mesh sizes (180, 315, and 500 μm) were chosen for sieving powders into granulometric classes so that each granulometric class represent a significant mass fraction of the unsieved powder.

3.2. Efficiency of okra seed powder sieving

Before proximate analysis and characterization of flow properties of the powders, the efficiency of the milling and sieving processes for the production of different granulometric classes were verified by weighing powder samples after sieving (Table 1).

Table 1

Table 1 shows that the sieving process allowed an efficient fractionation of okra seed powders into the different granulometric classes, as significant mass yields (generally above 10 %) were found for all sieved powders. As expected from particle size distributions of unsieved powders (figure 1), the $\geq 500 \mu\text{m}$ granulometric class was the most represented (around 50 % mass yield) whatever the milling speed. This is consistent with the fact that seeds are material of rather high hardness, making them difficult to grind, and resulting into powders containing a marked proportion of large particles.

3.3. Particle size distribution

Particle size distribution of all granulometric classes and unsieved powder are displayed in figures 2, 3, and 4 and associated granulometric parameters (d_{10} , d_{50} , d_{90} , and span) are presented in table 2.

Figure 2

Figure 3

Figure 4

Figures 2, 3, 4 and table 2 show that okra seed powders were correctly fractionated by sieving despite their rather large fat content (that will be discussed in the following subsection 3.4) likely to make them sticky. Indeed, particle size distributions of sieved powders were generally monomodal, with small span values (except for $\leq 180 \mu\text{m}$ granulometric classes whatever the milling speed and the 180 - 315 μm granulometric class at 12 000 rpm milling speed).

The large width of particle size distributions (span) of $\leq 180 \mu\text{m}$ granulometric classes whatever the milling speed and of the 180 - 315 μm granulometric class at 12 000 rpm milling speed may be explained by the presence of numerous fine particles (likely coming from the attrition of larger particles in the course of the milling process) that may adhere to larger ones and thus may be difficult to separate into individual particles. Indeed, as it will be evidenced in the next subsection (3.4) by proximate composition results, okra seed powders were rich in fat, which may favor powder cohesion and result in their agglomeration during the milling process, especially for fine particles, as they had a higher specific surface area and contained more fat.

Whatever it may be, the results of sieve fractionation (table 1) and particle size characteristics of powder fractions (table 2) showed that the successive milling and sieving procedure was effective in micronizing okra seeds and separating unsieved powders into sufficiently different size classes. Indeed, the d50 of sieved powders (granulometric classes) was systematically found between the mesh sizes of upper and lower sieves, which confirms the suitability of sieve shaking for separating okra seed powders.

Milling speed contributed to reduce mean particle size and span of granulometric classes (cf. figures 2, 3, and 4 and table 2), as expected. Both the decrease in particle sizes and the increase in the width of the particle size distribution at higher milling speed were consistent with the more elevated mass fractions of $\leq 180 \mu\text{m}$, $180 - 315 \mu\text{m}$, and $315 - 500 \mu\text{m}$ granulometric classes (table 1).

Table 2

3.4. Proximate composition

In table 3, contents in moisture, fat, proteins, ash, and carbohydrates of all granulometric classes and unsieved powders of okra seeds were evaluated to investigate the influence of milling speed and sieving efficiency. Indeed, proximate composition may be indicative of the stickiness of powder surface (for instance, surface fat generally tend to increase powder stickiness) (Langrish *et al.*, 2006; Viviane *et al.*, 2007), which is known to greatly impair powder flowability.

Table 3

Moisture content of granulometric classes $\leq 180 \mu\text{m}$, $180 - 315 \mu\text{m}$, and $315 - 500 \mu\text{m}$ was lower than 10 %, whereas unsieved powders and $\geq 500 \mu\text{m}$ samples had a moisture content ranging between 10 and 13 %. It is generally considered that a moisture content lower than 10 % ensures powder stability during storage (Kaur, Kaushal and Sandhu, 2013). Then, granulometric classes composed of smaller particles were expected to have better preservation abilities than unsieved powders and $\geq 500 \mu\text{m}$ samples. Also, fat and protein contents significantly decreased when the particle size was increased, unlike carbohydrates and ash that increased. As regards the influence of milling speed, it appeared that the contents in fat and proteins (resp., carbohydrates and ash) decreased (resp., increased) in $\leq 180 \mu\text{m}$ and $180 - 315 \mu\text{m}$ granulometric classes and increased (resp., decreased) in $315 - 500 \mu\text{m}$ and $\geq 500 \mu\text{m}$ granulometric classes. Indeed, it was expected that a more intense milling enhances the size reduction of plant material and thus the release of fat and proteins from plant cells (Becker *et al.*, 2016; Zaiter *et al.*, 2016). This can be related to okra seed structure: indeed, the outer shell

is composed of a higher proportion of fibers than other parts of okra seeds. Then, as fibers are more difficult to grind, larger particles may be enriched in this component, leading to higher carbohydrate content. Consequently, the proportions of fat and proteins were decreased in larger particles. Previous studies explained that plant parts containing a high proportion of fibers and sugars (both belonging to carbohydrates) are harder to grind and thus result in larger particles after milling, whereas plant parts with higher levels of fat and proteins are easier to grind and thus lead to smaller particles (Becker *et al.*, 2016; Zaiter *et al.*, 2016). So, proximate composition of okra seed powders was greatly dependent on granulometric class; in other words, the mean particle size had a marked impact on proximate composition of okra powders. The moisture content was decreased for powder samples constituted by small particles, because they probably undergo a higher friction-induced heating during milling. Moreover, statistical analysis permitted to show that large particles were richer in carbohydrates. Plant parts containing a large proportion of fibers are expected to be more difficult to grind and result in large particles. As fibers were included in total carbohydrates in the proximate composition analysis, the larger content in carbohydrates of higher size granulometric classes of okra seed powders may be due to a higher proportion of fibers. Bioactive compounds are often associated with proteic and fatty fractions, thus the smaller granulometric classes of okra seeds are expected to have higher contents in bioactive compounds (Becker, *et al* 2016; Deli, Ndjantou, *et al.*, 2019). At higher milling speed, this differential distribution of macronutrients within granulometric classes was enhanced: small particles contained even more proteins and fat and less moisture, carbohydrates, and ashes. Thus, the discriminating power of the sieving process in matter of proximate composition was improved at higher milling speed.

3.5. Flow properties of okra seed powders

3.5.1. Stability and compressibility tests

The results of stability tests for granulometric classes and unsieved samples of okra seed powders are presented in table 4.

Table 4

Whatever the milling speed, unsieved powders presented significant differences with granulometric classes in terms of BFE and SE. Moreover, the CBD of unsieved powders was systematically found within the range of CBD of granulometric classes. The CBD of granulometric classes decreased with particle size, indicating that the powder bed was less compact for samples composed of smaller particles. This could be an indication of higher

cohesion of smaller granulometric classes, which is consistent with their probable stickiness deriving from their higher fat content as well as with the fact that smaller particles are able to establish more contact points with their surrounding environment (particles, walls), thus impairing their flowability. At first glance, BFE results seemed in contradiction with this conclusion, as BFE decreased with mean particle size and this could be interpreted as better flowability of granulometric classes of smaller mean particle size. However, it should be mentioned that powder bed with poor flow properties may also lead to low BFE values when particle cohesion is so high that the blade only makes flow the small amount of powder located in its vicinity (Leturia *et al.*, 2014; Gnagne *et al.*, 2017). Also, interpretations of powder flowability on the basis of the BFE values may be distorted by important CBD differences. Indeed, the stability test is performed to evaluate the flowability of a given volume of conditioned powder bed, hence sample mass may vary between samples according to aerated density (i.e. CBD) differences. When comparing powder sample differing in CBD, the SE seems more appropriate to give information on powder flowability, as it corresponds to flow energy normalized against powder mass. All okra seed powders exhibited SE values close to 5 mJ/g, indicating that studied powders had correct flow properties. For samples milled at 6 000 rpm, the lower SE (indicative of better flowability) was found for the 180 - 315 μm granulometric class, while at 12 000 and 18 000 rpm milling speed, the best flow properties on the basis of the SE was found for the highest granulometric class. This is consistent with the fact that larger particles are expected to flow more easily (Petit *et al.*, 2017; Shi *et al.*, 2018). Also, the better flowability of the 180 - 315 μm granulometric class for samples milled at 6 000 rpm may result from the lower span of this sample (cf. table 2).

Moreover, the influence of milling speed on powder flowability can be deduced by comparing unsieved powders. Table 4 shows that the SE and the CBD increased, while the BFE decreased, when the milling speed was increased. This trend was expected, because when the milling speed increases, the mean particle size decreases and the interparticular interactions become so high that powders are much cohesive and have a poorer flowability.

Table 4 and figures 5, 6, and 7 present the results of the compressibility tests. Whatever the milling speed, compressibility markedly decreased with mean particle size, as expected: indeed, it has previously been discussed that smaller particles are generally more cohesive, making them more compressible. This confirms the better flow properties of powders composed of larger particles. Unsieved powders had intermediate compressibility, comprised in the range of compressibilities of granulometric classes. Compressibility of unsieved powders and

granulometric classes was increased at higher milling speed, consistently with the lower mean particle size, and this was expected to lower powder flowability.

Final densities after compression at 15 kPa CD_{15kPa} ranged between 0.5 and 0.7 g/mL for all powders, and these rather elevated values confirmed that the flowability of studied powders was between correct and good. All these interpretations of compressibility results were valid whatever the applied normal stress (cf. figures 5, 6, and 7).

Figure 5

Figure 6

Figure 7

3.5.2. Aeration and shear cell tests

Aeration results of granulometric classes and unsieved samples of okra seed powders are presented in table 5.

Table 5

All investigated okra seed powders were not fluidizable in standard conditions of the FT4 aeration test (air velocity up to 10 m.s⁻¹). Indeed, very low aeration ratios AR and elevated aerated energies AE were found, showing that the fluidized state was not achieved at maximal air velocity employed in the standard FT4 test. This may be explained by the high mean particle size of 180 - 315 µm, 315 - 500 µm, and ≥ 500 µm granulometric classes and the presumed relatively high cohesion of ≤ 180 µm and 180 - 315 µm granulometric classes (Gnagne et al., 2017; Petit et al., 2017). The low fluidizability of studied powders may constitute an advantage for handling purposes at industrial scale, as loss of these powders by projection in the surrounding air may be reduced. Aeration properties seemed not to be affected by changing the milling speed, which is logical, as the order of magnitude of mean particle size of powder samples was the same whatever the milling speed.

Shear test results are displayed in table 5 and figures 8, 9, and 10. Whatever the milling speed, the flow factor decreased when decreasing the mean particle size of granulometric classes, consistently with the increase in powder cohesion. Indeed, smaller particles are expected to be more cohesive (more interparticular contact points) and thus flow more poorly. Investigated powders ranged from cohesive to easy-flowing, according to Jenike classification (Jenike, 1964). Besides, the trends of decrease in flow factor and increase in cohesion at higher milling

speed were in agreement with the decreased mean particle size of unsieved powders. The results of the shear test met those of stability and compressibility tests, as powders with lower mean particle size needed more energy to flow and were more compressible. This permits to state that in our study the shear cell and compressibility tests were most suitable for sorting powders on the basis of their flowability.

Figure 8

Figure 9

Figure 10

4. Conclusion

The objective of this work was to determine the impact of particle size class of okra seed powders on their proximate composition and their flowability. Three different milling speeds were used to produce powders that were each separated in four granulometric sizes by sieving. It was found, as expected, that the mean particle size decreased when the milling speed was increased. The sieving process was successful in separating unsieved powders into sufficiently different size classes: a significant mass yield (generally above 10 %) was obtained for each sieved powder. Mean particle size was found to be comprised between the upper and lower sieves, confirming the efficiency of the sieving process. A strong correlation was found between mean particle size and proximate composition: more proteins and fat and less water, carbohydrates, and minerals were obtained for smaller granulometric classes. Also, the influence of milling speed and mean particle size of sieved powders on flow properties was denoted. Most samples were cohesive or easy-flowing. $\leq 180\mu\text{m}$, granulometric classes, having a lower mean particle size and a higher fat content, were significantly more compressible and cohesive than other samples. This information regarding the influence of milling speed and mean particle size on flow properties of okra seed powders should be useful for the production at industrial scale of okra seed powders, especially for handling, packaging, and storage purposes. A compromise should be found between content in nutritional quality and/or content in bioactive compounds, mass yields of the successive milling and sieving processes, and flow properties.

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6. References

- Abdullah, E. C. & Geldart, D. (1999). The use of bulk density measurements as flowability indicators. *Powder Technology*, **102**, 151–165.
- Adelakun, O.E., Oyelade, O.J., Ade-Omowaye, B.I.O., Adeyemi, I.A., M. Van de Venter. (2009). Mineral composition and the functional attributes of Nigerian okra seed (*Abelmoschus esculentus* Moench) flour. *Food and Chemical Toxicology*. Elsevier Ltd, **47**, 1123–1126. doi: 10.1016/j.foodres.2011.08.003.
- Adelakun, O. E. & Oyelade, O. J. (2011). Potential use of Okra Seed (*Abelmoschus Esculentus* Moench): Flour for Food Fortification and Effects of Processing. *Flour and Breads and their Fortification in Health and Disease Prevention*. Chapitre 19, 205-2012. doi: 10.1016/B978-0-12-380886-8.10019-4.
- ADPI (American Dry Product Institute, Elmhurst, IL). Determination of total ash in standards for grades of dry milks including methods of analysis, Bulletin 916' (2002). Annual Reviews, **40–41**.
- AFNOR (Agence Française de NORMALISATION), Détermination de la teneur en eau-méthode par étuvage' (1976), ((No: V04 – 348)).
- Becker, L., Zaiter, A., Petit, J., Zimmer, D., Karam M.C., Baudelaire, E., Scher, J., Dicko A., (2016). Improvement of antioxidant activity and polyphenol content of *Hypericum perforatum* and *Achillea millefolium* powders using successive grinding and sieving, *Industrial Crops and Products*. Elsevier B.V., **87**, 116–123. doi: 10.1016/j.indcrop.2016.04.036.
- Bradstreet, R. B. (1954). Kjeldahl Method for Organic Nitrogen', *Analytical Chemistry*, **26**, 185–187. doi: 10.1021/ac60085a028.
- Brewer, L. R. Kubola, J. Siriamornpun, S. Herald T, J. Shi, Y. C. (2014). Wheat bran particle size influence on phytochemical extractability and antioxidant properties. *Food Chemistry*. Elsevier Ltd, **152**, 483–490. doi: 10.1016/j.foodchem.2013.11.128.
- Camciuc, M., Deplagne, M., Vilarem, G. Gaset, A., (1998). Okra - *Abelmoschus esculentus* L. (Moench.) a crop with economic potential for set aside acreage in France. *Industrial Crops and Products*. Elsevier, **7**, 257–264. doi: 10.1016/S0926-6690(97)00056-3.
- Castro, M. D. & Garcia-Ayuso, L. E. (1998). Soxhlet extraction of solid materials : an outdated technique with a promising innovative future. *ANALYTICA CHIMICA ACTA*, **369**, 1–10.
- Chen, Y., Zhang, B. C., Sun, Y. H., Zhang, J. G., Sun, H. J., Wei, Z. J., (2015). Physicochemical properties and adsorption of cholesterol by okra (*Abelmoschus esculentus*) powder. *Food and Function*. Royal Society of Chemistry, **6**, 3728–3736. doi: 10.1039/c5fo00600g.
- Crayveld, V., Holopainen, U., Selinheima, E., Kaisa, P., Delcour, J. A., Courtin, C. M. (2009). Extensive Dry Ball Milling of Wheat and Rye Bran Leads to in Situ Production of Arabinoxylan Oligosaccharides through Nanoscale Fragmentation. *Journal of Agricultural and Food Chemistry*, **57**, 8467–8473. doi: 10.1021/jf901870r.
- Deli, M., Petit, J., Nguimbou, R. M., Beaudelaire, E., Njintang, Y. N., Scher, J. (2019a). Effect of sieved fractionation on the physical, flow and hydration properties of *Boscia senegalensis* Lam., *Dichostachys glomerata* Forssk. and *Hibiscus sabdariffa* L. powders. *Food Science and*

- 489 *Biotechnology*. Springer Singapore, **22**, 1–15. doi: 10.1007/s10068-019-00597-6.
- 490 Deli, M., Baudelaire, E., Metsagang, J.T., N. Petit, J., Njintang, Y. N., Scher, J. (2019b).
 491 Successive grinding and sieving as a new tool to fractionate polyphenols and antioxidants of
 492 plants powders: Application to *Boscia senegalensis* seeds, *Dichrostachys glomerata* fruits, and
 493 *Hibiscus sabdariffa* calyx powders. *Food Science and Nutrition*, **7**, 1795–1806. doi:
 494 10.1002/fsn3.1022.
- 495 Freeman, R. (2007). Measuring the flow properties of consolidated , conditioned and aerated
 496 powders - A comparative study using a powder rheometer and a rotational shear cell. *Powder*
 497 *Technology*, **174**, 25–33. doi: 10.1016/j.powtec.2006.10.016.
- 498 Fu, X., Huck, D., Makein, L., Armstrong, B., Willen, U., Freeman, T. (2012). Effect of particle
 499 shape and size on flow properties of lactose powders. *Particuology*. Chinese Society of
 500 Particuology, **10**, 203–208. doi: 10.1016/j.partic.2011.11.003.
- 501 Gnagne, E. H., Petit, J., Gaiani, C., Scher, J., Amani, G. N., (2017). Characterisation of flow
 502 properties of foutou and fofou flours, staple foods in West Africa, using the FT4 powder
 503 rheometer. *Journal of Food Measurement and Characterization*. Springer US, **11**, pp. 1128–
 504 1136. doi: 10.1007/s11694-017-9489-2.
- 505 Jarret, R. L., Wang, M. L., & Levy, I. J. (2011). Seed oil and fatty acid content in okra
 506 (*Abelmoschus esculentus*) and related species. *Journal of Agricultural and Food Chemistry*,
 507 **59**, 4019–4024. doi: 10.1021/jf104590u.
- 508 Jenike, A. W. (1964). Storage and Flow of Solids. *Bulletin of the University of Utah*, **53**, p. 209.
 509 doi: 10.2172/5240257.
- 510 Kaur, M., Kaushal, P., and Sandhu, K. S., (2013). Studies on physicochemical and pasting
 511 properties of Taro (*Colocasia esculenta* L.) flour in comparison with a cereal, tuber and legume
 512 flour. *Journal of Food Science and Technology*, **50**, pp. 94–100. doi: 10.1007/s13197-010-
 513 0227-6.
- 514 Langrish, T. A. G., Marquez, N., & Kota, K. (2006). An investigation and quantitative
 515 assessment of particle shape in milk powders from a laboratory-scale spray dryer. *Drying*
 516 *Technology*, **24**, 1619–1630. doi: 10.1080/07373930601031133.
- 517 Leturia, M., Benali, M., Lagarde, S., Ronga, I., Saleh, K. (2014). Characterization of fl ow
 518 properties of cohesive powders : A comparative study of traditional and new testing methods.
 519 *Powder Technology*. Elsevier B.V., **253**, 406–423. doi: 10.1016/j.powtec.2013.11.045.
- 520 Lucas-González, R., Viuda-Martos, M., Pérez-Álvarez, J. Á., Fernández-López, J., (2017).
 521 Evaluation of Particle Size Influence on Proximate Composition, Physicochemical, Techno-
 522 Functional and Physio-Functional Properties of Flours Obtained from Persimmon (*Diospyros*
 523 *kaki* Trumb.) Coproducts. *Plant Foods for Human Nutrition*. **72**, 67–73. doi: 10.1007/s11130-
 524 016-0592-z.
- 525 Miller, J.C. Wilson, W.F & Meadows, M. (1949). Growing okra seed for its oil. *Chemurg. Dig*,
 526 **8**, 22–24.
- 527 Ndangui, C. B. Kimbonguila, A. Nzikou, J. M. Matos, L. Pambou-Tobi, N. P. G. Abena, A. A.
 528 Silou, Th. Scher, J. Desobry, S. (2010). Nutritive Composition and Properties Physico-chemical
 529 of gumbo (*Abelmoschus esculentus* L.) Seed and Oil. *Research Journal of Environmental and*
 530 *Earth Sciences*, **2**, 49–54.
- 531 Oyelade, O. J., Ade-Omowaye, B. I. O., and Adeomi, V. F. (2003). Influence of variety on

- protein, fat contents and some physical characteristics of okra seeds. *Journal of Food Engineering*, **57**, 111–114. doi: 10.1016/S0260-8774(02)00279-0.
- Petit, J., Burgain, J., Gainani, C. & Scher, J. (2017). Aptitude à l'écoulement de poudres alimentaires: impact des propriétés physicochimiques des particules. *Industries alimentaires et agricoles (IAA)*, pp. 26–30.
- Shi, H., Mohanty, R., Chakravarty, S., Cabisco, R., Morgeneyer, M., Zetzener, H. O., Jin Y., Kwade, A., Luding, S., Magnanimo, V. (2018). Effect of particle size and cohesion on powder yielding and flow. *KONA Powder and Particle Journal*, **14**, 226–250. doi: 10.14356/kona.2018014.
- Torkpo, S., Danquah, E.Y., Offei, S. K., Blay, E.T. (2009). Esterase, total protein and seed storage protein diversity in Okra (*Abelmoschus esculentus* L. Moench). *West African Journal of Applied Ecology*, **9**, 1–7. doi: 10.4314/wajae.v9i1.45677.
- Uka, U., Chukwuka, K. & Iwuagwu, M. (2013). Relative effect of organic and inorganic fertilizers on the growth of okra [*Abelmoschus esculentus* (L.) Moench]. *Journal of Agricultural Sciences, Belgrade*, **58**, 159–166. doi: 10.2298/jas1303159u.
- Viviane, S., Birchall, M., Laura, P., Glória, R. S., Wildhagen, A. S., Mujumdar. (2007). Effect of Spray-Dryer Operating Variables on the Whole Milk Powder Quality. *Drying Technology*, **23**, 611–636. doi: 10.1081/DRT-200054153.
- Yu, W., Muteki, K., Zhang, L., & Kim, G. (2011). Prediction of bulk powder flow performance using comprehensive particle size and particle shape distributions. *Journal of Pharmaceutical Sciences*. Elsevier Masson SAS, **100**, 284–293. doi: 10.1002/jps.22254.
- Zaiter, A., Becker, L., Petit, J., Zimmer, D., Karam, M. C., Baudelaire, E., Scher, J., Dicko, A.. (2016). Antioxidant and antiacetylcholinesterase activities of different granulometric classes of *Salix alba* (L.) bark powders. *Powder Technology*. Elsevier B.V., **301**, 649–656. doi: 10.1016/j.powtec.2016.07.014.
- Zhang, Z., Song, H., Peng, Z., Luo, Q., Ming, J., Zhao, G. (2012). Characterization of stipe and cap powders of mushroom (*Lentinus edodes*) prepared by different grinding methods. *Journal of Food Engineering*. Elsevier Ltd, **109**, 406–413. doi: 10.1016/j.jfoodeng.2011.11.007.
- Zhao, X., Yang, Z., Gai, G., Yang, Y. (2009). Effect of superfine grinding on properties of ginger powder. *Journal of Food Engineering*. Elsevier Ltd, **91**, 217–222. doi: 10.1016/j.jfoodeng.2008.08.024.

Figure 1: Particle size distributions of okra seed unsieved powders obtained at 6 000, 12 000, and 18 000 rpm milling speed.

Figure 2: Particle size distributions of the different granulometric classes and unsieved sample of okra seed powders obtained by milling at 6 000 rpm.

Figure 3: Particle size distributions of the different granulometric classes and unsieved sample of okra seed powders obtained by milling at 12 000 rpm.

Figure 4: Particle size distributions of the different granulometric classes and unsieved sample of okra seed powders obtained by milling at 18 000 rpm.

Figure 5: Compressibility of the different granulometric classes and unsieved sample of okra seed powders obtained by milling at 6 000 rpm. Error bars represent standard deviations; some were smaller than the marker size and thus not visible.

Figure 6: Compressibility of the different granulometric classes and unsieved sample of okra seed powders obtained by milling at 12 000 rpm. Error bars represent standard deviations; some were smaller than the marker size and thus not visible.

Figure 7: Compressibility of the different granulometric classes and unsieved sample of okra seed powders obtained by milling at 18 000 rpm. Error bars represent standard deviations; some were smaller than the marker size and thus not visible.

Figure 8: Shear test curve obtained after pre-shear at 9 kPa normal stress for the different granulometric classes and unsieved sample of okra seed powders obtained by milling at 6 000 rpm. Error bars represent standard deviations; some were smaller than the marker size and thus not visible.

Figure 9: Shear test curve obtained after pre-shear at 9 kPa normal stress for the different granulometric classes and unsieved sample of okra seed powders obtained by milling at 12 000 rpm. Error bars represent standard deviations; some were smaller than the marker size and thus not visible.

Figure 10: Shear test curve obtained after pre-shear at 9 kPa normal stress for the different granulometric classes and unsieved sample of okra seed powders obtained by milling at 18 000 rpm. Error bars represent standard deviations; some were smaller than the marker size and thus not visible.

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Table 1: Sieved masses and mass fractions of the different granulometric classes and unsieved powders of okra seed powders obtained at 6 000, 12 000, and 18 000 rpm milling speed.

Milling speed (rpm)	6 000		12 000		18 000	
Granulometric class	Sieved mass (g)	Mass yield (%)	Sieved mass (g)	Mass yield (%)	Sieved mass (g)	Mass yield (%)
≥ 500 µm	317.53	59.13	164.18	50.26	86.83	42.47
315 - 500 µm	101.05	18.82	52.34	16.02	41.75	20.42
180 - 315 µm	51.16	9.53	44.68	13.68	46.32	22.66
≤ 180 µm	67.26	12.52	65.44	20.04	29.54	14.45
Unsieved powder (total)	537.00	100.00	326.64	100.00	204.44	100.00

Table 2: Granulometric parameters of the different granulometric classes and unsieved samples of okra seed powders obtained at 6 000, 12 000, and 18 000 rpm milling speed.

Milling speed (rpm)	6 000			
Granulometric classes	d10 (µm)	d50 (µm)	d90 (µm)	Span (-)
≥ 500 µm	589.33 ± 18.50 ^d	1100.00 ± 43.59 ^e	2173.33 ± 73.71 ^e	1.44 ± 0.02 ^c
315 - 500 µm	326.67 ± 6.03 ^c	498.33 ± 2.52 ^c	745.33 ± 2.52 ^c	0.87 ± 0.03 ^a
180 - 315 µm	152.00 ± 1.00 ^b	257.00 ± 1.00 ^b	406.00 ± 1.00 ^b	0.97 ± 0.01 ^b
≤ 180 µm	23.10 ± 0.20 ^a	85.30 ± 0.61 ^a	181.33 ± 0.58 ^a	1.87 ± 0.01 ^d
Unsieved powder	141.00 ± 7.81 ^b	723.33 ± 29.96 ^d	1670.00 ± 65.57 ^d	2.11 ± 0.02 ^e
Milling speed (rpm)	12 000			
Granulometric classes	d10 (µm)	d50 (µm)	d90 (µm)	Span (-)
≥ 500 µm	542.67 ± 5.69 ^e	801.33 ± 25.02 ^d	1243.33 ± 75.06 ^e	0.90 ± 0.05 ^b
315 - 500 µm	312.00 ± 1.00 ^d	463.33 ± 1.15 ^c	671.67 ± 4.93 ^c	0.78 ± 0.01 ^a
180 - 315 µm	165.67 ± 1.53 ^c	264.67 ± 1.53 ^b	409.33 ± 9.81 ^b	0.92 ± 0.03 ^b
≤ 180 µm	21.90 ± 0.10 ^a	75.60 ± 2.84 ^a	167.33 ± 6.35 ^a	1.95 ± 0.06 ^c
Unsieved powder	52.93 ± 0.58 ^b	457.67 ± 5.13 ^c	983.00 ± 7.81 ^d	2.03 ± 0.01 ^c
Milling speed (rpm)	18 000			
Granulometric classes	d10 (µm)	d50 (µm)	d90 (µm)	Span (-)
≥ 500 µm	527.67 ± 4.73 ^e	781.00 ± 14.18 ^e	1186.67 ± 37.86 ^d	0.84 ± 0.02 ^b
315 - 500 µm	290.00 ± 4.36 ^d	442.67 ± 12.86 ^d	644.67 ± 25.93 ^c	0.80 ± 0.02 ^b
180 - 315 µm	37.40 ± 0.17 ^c	181.00 ± 0.00 ^b	332.67 ± 2.08 ^b	0.48 ± 0.01 ^a
≤ 180 µm	17.83 ± 0.25 ^a	52.13 ± 0.65 ^a	149.33 ± 1.53 ^a	0.77 ± 0.01 ^b
Unsieved powder	25.70 ± 0.36 ^b	228.67 ± 7.51 ^c	702.67 ± 55.77 ^c	2.96 ± 0.16 ^c

For powder results at each milling speed separately, means ± standard deviations followed by the same superscripted letters in the same column were not significantly different at $p < 0.05$ according to Tukey's HSD test.

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Table 3: Proximate composition (contents in moisture, lipids, proteins, ash, carbohydrates on wet basis) of the different granulometric classes and unsieved powders of okra seeds.

Milling speed (rpm)	6 000					
Powder sample	Unsieved powder	≤ 180 μm	180 - 315 μm	315 - 500 μm	≥ 500 μm	Unsieved powder [3]
Moisture %(w/w))	11.10 ± 0.07 ^b	9.59 ± 0.12 ^a	9.01 ± 0.67 ^a	9.21 ± 0.11 ^a	12.07 ± 0.05 ^c	9.45 ± 0.80
Fat %(w/w))	12.05 ± 0.07 ^b	23.61 ± 0.01 ^c	23.14 ± 0.43 ^c	23.34 ± 0.02 ^c	7.63 ± 0.20 ^a	24.90 ± 0.55
Proteins %(w/w))	16.45 ± 0.74 ^b	32.08 ± 0.81 ^d	30.07 ± 1.19 ^{cd}	28.77 ± 0.29 ^c	10.77 ± 0.62 ^a	24.85 ± 1.50
Carbohydrates %(w/w))	55.44 ± 0.82 ^b	28.33 ± 0.77 ^a	30.28 ± 1.63 ^a	32.12 ± 0.42 ^a	68.70 ± 8.04 ^c	36.83 ± 1.78
Ash %(w/w))	4.96 ± 0.04 ^a	6.39 ± 0.01 ^b	7.00 ± 0.66 ^b	6.56 ± 0.07 ^b	4.21 ± 0.46 ^a	5.68 ± 0.10
Milling speed (rpm)	12 000					
Powder sample	Unsieved powder	≤ 180 μm	180 - 315 μm	315 - 500 μm	≥ 500 μm	Unsieved powder [3]
Moisture %(w/w))	11.27 ± 0.08 ^c	9.15 ± 0.10 ^b	8.61 ± 0.08 ^a	9.22 ± 0.05 ^b	11.79 ± 0.06 ^d	9.45 ± 0.80
Fat %(w/w))	13.71 ± 0.89 ^b	26.69 ± 0.54 ^e	23.40 ± 0.27 ^d	19.21 ± 0.22 ^c	5.18 ± 0.17 ^a	24.90 ± 0.55
Proteins %(w/w))	17.04 ± 0.31 ^b	34.35 ± 0.62 ^e	30.61 ± 0.27 ^d	27.63 ± 1.12 ^c	7.84 ± 0.30 ^a	24.85 ± 1.50
Carbohydrates %(w/w))	51.01 ± 0.35 ^d	18.79 ± 0.68 ^a	30.53 ± 0.20 ^b	37.58 ± 1.19 ^c	70.33 ± 0.35 ^e	36.83 ± 1.78
Ash %(w/w))	5.12 ± 0.15 ^b	6.73 ± 0.04 ^d	6.23 ± 0.04 ^c	5.83 ± 0.09 ^c	4.09 ± 0.35 ^a	5.68 ± 0.10
Milling speed (rpm)	18 000					
Powder sample	Unsieved powder	≤ 180 μm	180 - 315 μm	315 - 500 μm	≥ 500 μm	Unsieved powder [3]
Moisture %(w/w))	10.69 ± 0.09 ^c	9.56 ± 0.08 ^a	10.15 ± 0.02 ^b	11.32 ± 0.06 ^d	13.20 ± 0.17 ^e	9.45 ± 0.80
Fat %(w/w))	14.57 ± 0.69 ^c	27.80 ± 0.51 ^e	23.51 ± 0.08 ^d	12.17 ± 0.15 ^b	3.43 ± 0.43 ^a	24.90 ± 0.55
Proteins %(w/w))	18.37 ± 0.12 ^b	35.01 ± 0.26 ^e	28.62 ± 0.24 ^d	19.72 ± 0.72 ^c	5.83 ± 0.53 ^a	24.85 ± 1.50
Carbohydrates %(w/w))	50.99 ± 1.23 ^c	24.73 ± 0.68 ^a	33.33 ± 0.88 ^b	52.94 ± 2.52 ^c	73.19 ± 0.76 ^d	36.83 ± 1.78
Ash %(w/w))	5.18 ± 0.04 ^c	6.80 ± 0.01 ^e	5.94 ± 0.06 ^d	4.39 ± 0.16 ^b	3.75 ± 0.28 ^a	5.68 ± 0.10

Table 4: Results of stability and compressibility tests for unsieved samples and granulometric classes of okra seed powders produced at 6 000, 12 000, and 18 000 rpm milling speed.

Milling speed (rpm)	6 000				
Powder sample	Unsieved powder	$\leq 180 \mu\text{m}$	180 - 315 μm	315 - 500 μm	$\geq 500 \mu\text{m}$
BFE (mJ)	285.00 ± 2.27^d	93.12 ± 2.00^a	164.96 ± 10.12^b	197.49 ± 14.34^c	265.58 ± 7.92^d
SE (mJ/g)	6.70 ± 0.24^c	5.94 ± 0.14^{bc}	4.64 ± 0.61^a	4.91 ± 0.31^{ab}	5.50 ± 0.50^{ab}
CBD (g/mL)	0.53 ± 0.02^d	0.33 ± 0.01^a	0.44 ± 0.00^b	0.50 ± 0.01^c	0.58 ± 0.00^c
CD _{15kPa} (g/mL)	0.63 ± 0.02^d	0.50 ± 0.01^a	0.53 ± 0.00^b	0.57 ± 0.02^c	0.64 ± 0.00^d
CP _{15kPa} (%)	15.30 ± 0.67^c	33.34 ± 1.09^d	16.95 ± 0.00^c	11.10 ± 0.50^b	8.43 ± 0.28^a
Milling speed (rpm)	12 000				
Powder sample	Unsieved powder	$\leq 180 \mu\text{m}$	180 - 315 μm	315 - 500 μm	$\geq 500 \mu\text{m}$
BFE (mJ)	259.04 ± 6.43^c	89.88 ± 7.19^a	190.37 ± 2.47^b	252.10 ± 6.53^c	248.57 ± 7.03^c
SE (mJ/g)	7.08 ± 0.13^c	7.08 ± 0.11^c	5.45 ± 0.07^b	5.56 ± 0.23^b	4.50 ± 0.61^a
CBD (g/mL)	0.53 ± 0.01^c	0.33 ± 0.03^a	0.42 ± 0.01^b	0.50 ± 0.01^c	0.61 ± 0.02^d
CD _{15kPa} (g/mL)	0.67 ± 0.01^b	0.57 ± 0.06^a	0.53 ± 0.00^a	0.57 ± 0.01^a	0.66 ± 0.02^b
CP _{15kPa} (%)	21.98 ± 0.18^c	42.01 ± 0.89^d	20.92 ± 0.62^c	13.75 ± 0.58^b	7.56 ± 0.11^a
Milling speed (rpm)	18 000				
Powder sample	Unsieved powder	$\leq 180 \mu\text{m}$	180 - 315 μm	315 - 500 μm	$\geq 500 \mu\text{m}$
BFE (mJ)	219.48 ± 3.48^b	88.09 ± 17.89^a	204.28 ± 17.54^b	311.58 ± 7.16^c	299.17 ± 21.76^c
SE (mJ/g)	7.12 ± 0.41^b	6.88 ± 0.61^b	6.49 ± 0.19^{ab}	6.14 ± 0.21^{ab}	5.68 ± 0.22^a
CBD (g/mL)	0.47 ± 0.00^c	0.33 ± 0.03^a	0.42 ± 0.01^b	0.51 ± 0.01^d	0.60 ± 0.01^c
CD _{15kPa} (g/mL)	0.66 ± 0.01^b	0.56 ± 0.04^a	0.56 ± 0.01^a	0.59 ± 0.01^a	0.65 ± 0.02^b
CP _{15kPa} (%)	29.29 ± 0.74^d	41.44 ± 1.77^c	24.35 ± 0.42^c	12.66 ± 0.16^b	7.31 ± 0.39^a

Means $\pm$ standard deviations followed by the same superscripted letters in the same row were not significantly different at $p < 0.05$ according to Tukey's HSD test.

Table 5: Aeration and shear cell test results for unsieved powders and granulometric classes of okra seed powders obtained at 6 000, 12 000, and 18 000 rpm milling speed.

Milling speed (rpm)	6 000				
Powder sample	Unsieved powder	≤ 180 μm	180 - 315 μm	315 - 500 μm	≥ 500 μm
Aerated energy AE (mJ)	85.77 ± 13.53 ^b	21.12 ± 2.56 ^a	44.26 ± 2.40 ^a	51.45 ± 12.88 ^a	108.48 ± 20.79 ^b
Aeration ratio AR (-)	1.25 ± 0.07 ^a	2.53 ± 0.19 ^b	1.39 ± 0.13 ^a	1.16 ± 0.09 ^a	1.12 ± 0.10 ^a
Cohesion (kPa)	0.84 ± 0.09 ^a	1.88 ± 0.18 ^b	1.12 ± 0.19 ^a	0.92 ± 0.08 ^a	0.81 ± 0.10 ^a
Flow factor ffc (-)	5.29 ± 0.53 ^c	2.41 ± 0.16 ^a	3.91 ± 0.62 ^b	4.88 ± 0.42 ^{bc}	5.32 ± 0.59 ^c
Milling speed (rpm)	12 000				
Powder sample	Unsieved powder	≤ 180 μm	180 - 315 μm	315 - 500 μm	≥ 500 μm
Aerated energy AE (mJ)	104.87 ± 26.69 ^a	58.21 ± 25.21 ^a	73.12 ± 9.82 ^a	80.95 ± 11.52 ^a	64.62 ± 1.39 ^a
Aeration ratio AR (-)	1.53 ± 0.15 ^b	2.14 ± 0.23 ^c	1.26 ± 0.03 ^{ab}	1.06 ± 0.01 ^a	0.99 ± 0.08 ^a
Cohesion (kPa)	1.45 ± 0.22 ^b	3.05 ± 0.19 ^c	1.39 ± 0.32 ^b	0.96 ± 0.15 ^{ab}	0.69 ± 0.18 ^a
Flow factor ffc (-)	3.14 ± 0.59 ^{ab}	1.60 ± 0.09 ^a	3.34 ± 0.74 ^{ab}	4.65 ± 0.82 ^{bc}	6.44 ± 2.42 ^c
Milling speed (rpm)	18 000				
Powder sample	Unsieved powder	≤ 180 μm	180 - 315 μm	315 - 500 μm	≥ 500 μm
Aerated energy AE (mJ)	75.25 ± 1.37 ^b	31.91 ± 3.95 ^a	56.05 ± 18.34 ^{ab}	76.50 ± 8.16 ^b	61.15 ± 5.81 ^b
Aeration ratio AR (-)	1.47 ± 0.04 ^a	2.09 ± 0.13 ^b	1.38 ± 0.30 ^a	1.18 ± 0.03 ^a	1.09 ± 0.20 ^a
Cohesion (kPa)	1.64 ± 0.26 ^b	2.61 ± 0.20 ^c	1.48 ± 0.17 ^b	0.76 ± 0.22 ^a	0.48 ± 0.11 ^a
Flow factor ffc (-)	2.80 ± 0.40 ^a	1.80 ± 0.05 ^a	3.06 ± 0.28 ^a	6.11 ± 1.24 ^b	8.98 ± 2.09 ^b

Means ± standard deviations followed by the same superscripted letters in the same row were not significantly different at p < 0.05 according to Tukey’s HSD test.

Conflict of interests:

The authors declare that they have no conflict of interest.

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Ethics statements:

No experiment was performed on humans or animals. So, ethics approval was not required for this research.

For Peer Review

Data Availability Statement:

Research data are not shared.

For Peer Review

The authors wish to thank the reviewers for their new helpful comments. Detailed point-by-point response can be found hereafter, with reviewer comments in black (line numbers associated with reviewer comments correspond to submitted manuscript) and answers in blue (line numbers related to manuscript revision correspond to revised manuscript). In revised manuscript, additions were performed in blue and deletions in red crossed words.

Reviewer #1

The revision is satisfactory.

Thank you for your work in reviewing the manuscript.

Reviewer #2

Although the work presents interesting, it is quite hard to understand the paper due to poor communication. therefore i couldn't identify the main goal of this paper.Overall, this is not a well-written manuscript. There were no sufficient information about the previous study findings presented for readers to follow the present study rationale and procedures. Finally, I recommend that the paper should not be accepted for the publication in the present from.

Thank you for your work in reviewing the manuscript, the authors have tried to take into account these remarks in the second revised manuscript.

- L47: this sentence is not required it is repeating

The sentence in l41-43 indicates that okra (in general, i.e. all plant parts being considered) provides carbohydrates, protein, fat, minerals, and vitamins that are usually deficient in staple foods. The sentence in l47 concerns more specifically okra seeds, which is the studied material. Thus, it was decided to keep this sentence giving further information about the composition of okra seeds.

L49-50: Seeds are rich in proteins (45%) and lipids (34%), thus use as a food for a malnutrition children in Nigeria. (reference).

In order to reduce the length of this sentence, the authors preferred to delete "classifying is as a nutrient-rich plant". Also, "A research program initiated by Nigeria" was corrected by "A Nigerian research program".

- L52-54: rewrite, English is not good

This sentence was rewritten (cf. I51-53).

- L56: this sentence is not clear

Indeed, this convoluted sentence was removed.

- L58: you should discuss about this before you discuss the aim of this paper? you need to explain why you need to study this and the importance of this area and its relationship to human health as well as to okra

The link between nutritional quality and granulometric class has been added in I55-61, which permits to better introduce the aim of the study. The link with human health (food supplements) was added in I64-65.

- L59: this sentence and previous sentence has a big jump

"flow properties" was removed as it was not introduced previously. The link with the distribution of nutritional compounds within granulometric classes was made through the production of food supplements (cf. I64-65). The reason for studying also flow properties has been developed in I74-79.

- L64: not clear why

Indeed, this should now be clearer with the mention of "food supplements" in I64-65.

- L66: I what are the bioactive compounds you are going to extract not clear.

Indeed, this was out of the scope of the study, and "bioactive" as "replaced" by "nutritional" (cf. I67-68).

- L67: if you introduce new cofounders you need to explain them.

The authors did not understand this comment, nevertheless this sentence was out of the scope of the study and it was removed.

L68: I can't understand your English.

This sentence was deleted.

L70: not clear

The authors did not understand what is not clear in this sentence, and thus it as kept unchanged.

L73: not clear

This sentence was corrected in I83-84.

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L78: your introduction and the purpose is not matching rewrite.
With changes made in the second revised manuscript, the authors hope that the link between bibliographical data and study aim is now clearer.

L92-93: rewrite
The authors did not understand why this sentence should be rewritten, so it was kept unchanged.

L94: it is important do proof reading of your paper
This sentence appears clear enough, so the authors do not understand why this comment was formulated by reviewer #2. Maybe the fact that the sentence was justified and not aligned to the left? This was changed in l105.

L98: this sentence is too long rewrite
The authors did not find that this sentence is too long (42 words, 245 characters including spaces), and only one idea is presented in this sentence, so it was decided to keep this sentence unchanged.

L101: what do you mean by issued
This was corrected by "resulting from"

L104: you English is very poor so hard to understand.
The authors really did not understand this comment, as concerned sentence cannot be clearer.

L225-226: I can't understand what you mean by two populations.
It is very classical to talk about particle populations when discussing particle size distribution results. A particle population gathers all particles having similar characteristics, like size in the case of granulometric analysis. See here for instance: http://www-odp.tamu.edu/publications/178_SR/Volume/CHAPTERS/SR178_24.PDF