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Original Article

Morphological, textural analysis and freeze-thaw stability of starches from legume grow in Cameroon

Análise morfológica, textural e de estabilidade de congelamento-descongelamento de amidos de leguminosas cultivadas em Camarões

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Abstract

Starches from some legume grown in Cameroon were evaluated for their granule structure and size, turbidity, firmness and gel strength, thermal and freeze-thaw properties. Amylose contents were in the range of 26.21%–44.85%. Morphological analysis of the starch granules showed bimodal distribution, multiple sizes and shapes from small spherical to the bigger kidney shape. Significant differences were observed among starch in light transmittance, firmness and gel strength. The thermal parameters of starches were evaluated using differential scanning calorimeter and significant differences were observed. The peak gelatinisation temperature was positively correlated to starch granule size but the amylose content showed no evidence of their impact on legume starch properties studied. The data reported can be useful to facilitate the selection of variety of legume and conditions closer to the desired application.

Keywords: legume, starch, gelatinization, syneresis, freeze-thaw cycles.

Resumo

Neste trabalho, amidos de algumas leguminosas cultivadas em Camarões foram avaliados quanto à estrutura e tamanho dos grânulos, turbidez, firmeza e resistência do gel, propriedades térmicas e de congelamento e descongelamento. Os conteúdos de amilose estavam na faixa de 26,21% a 44,85%. A análise morfológica dos grânulos de amido mostrou distribuição bimodal, múltiplos tamanhos e formas desde o pequeno esférico até o maior formato de rim. Diferenças significativas foram observadas entre os amidos na transmissão de luz, firmeza e força do gel. Os parâmetros térmicos dos amidos foram avaliados usando calorímetro diferencial de varredura e diferenças significativas foram examinadas. A temperatura de pico de gelatinização foi positivamente correlacionada com o tamanho do grânulo de amido, no entanto, o teor de amilose não mostrou evidência de impacto nas propriedades do amido de leguminosas estudadas. Os dados informados podem ser úteis para facilitar a seleção de variedades de leguminosas e condições mais próximas da aplicação desejada.

Palavras-chave: leguminosa, amido, gelatinização, sinérese, ciclos de congelamento-descongelamento.

1. Introduction

Starch is a carbohydrate consisting of a large number of glucose units joined together by glycosidic bonds. This polysaccharide is produced by all green plants as an energy store. It is the most common carbohydrate in the human diet. It is composed of mainly amylose and amylopectin in different ratios. These two starch components account for approximately 98 to 99% of starch on the dry weight basis. Starch constitutes a major energy supply for humans worldwide and is produced as a reserve carbohydrate in plants. The most important sources for humans are diverse cereals, rhizomes, roots and tubers. Storage starch is produced in amyloplasts as discrete granules with distinct morphology in different plants, ranging from round, oval, ogival or elongated to flat, lenticular or polyhedral, and sizes from sub-microns to more than 100 μm in diameter (Bertoft, 2017). Although a great number of vegetables present considerable starch contents, only a few of these sources have been commercially explored, from which corn, wheat, rice, potato and cassava stand out. The food industry is the greatest starch consumer, but other industries, especially chemical and textile industries, also use this polymer. Research on new starchy raw materials has intensified in recent years, due mainly to restrictions imposed by the food industries on the use of modified starches. Starch is modified physically or chemically in order to obtain heat stable or resistance towards heat product, stable during freeze-thaw

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process and easily dissolve either in hot or cold suspension (Syahariza and Yong, 2017). Precedent study on using chemically modified starch in replacing fat in yogurt-based products for example, suggested a good effect in syneresis and good flow and better viscoelastic characteristics (Lobato-Calleros et al., 2014; Wang et al., 2019). However, there are two major disadvantages of utilizing solely pure starch which first is high attraction towards water and second is extreme rigidity. The disadvantages of using chemical modification are easily causing chemical residue to be left after modifying starch and definitely not an eco-friendly even though it is a fast, simple, extensively used and efficient method (Li et al., 2018). Food industry favors starch which is stable, white in color and odorless thus making legume seed as a perfect candidate by having around 60% starch. It has been shown recently that some legume seed like Bambara groundnut (Vigna subterranea) was an underutilized food crops in Africa and currently explored of its potential in terms of starch isolation (Oyeyinka et al., 2021). It is crucial to explore new and underutilized starch as it offers multiple functions in food and non-food industry. Thus, throughout the world, starch producing companies are increasing their search for natural starches with characteristics that attend the interests of the different types of industries, with special attention given to the food industry (Li et al., 2019). Alternative starch sources are being studied for both scientific and economic reasons. The determination of the proximate composition of starch and a microscopic examination are of fundamental importance in characterizing the susceptibility of the starch to a determined technological treatment. Therefore, it is possible to relate morphological and chemical characteristics to the physical and rheological properties of the products obtained from a determined starch (Ferreira-Villadiegoa et al., 2018) Starches from different sources can be characterized according to the different shapes and sizes of their granules. Our present investigation was undertaken to study and compare morphological, thermal, and pasting properties of starches from eight varieties of bean cultivars grown in Cameroon. Some information about processing properties of those varieties of beans including gel strength, firmness and syneresis have been identified. Finally, an attempt has been made to correlate the various starch properties and processing.

2. Material and Methods

2.1. Material

Three species of dry legume seeds including six varieties of *Phaseolus vulgaris* L. (Common bean: red bean, white bean, green bean, black bean, red spotted bean and yellow bean) one variety of *Vigna unguiculata* L. (Cowpea) and one variety of *Voandzeia subterranean* V. (Bambara groundnut) were used in this study. They were purchased from a local market in the Adamawa and West Regions of Cameroon in the Central Africa. The dry seeds of each variety were separately cleaned, uniformed in size and color, and freed from foreign or abnormal seeds and living or dead insects. For each variety, 3 kg portions of breeding seeds were placed in an aluminum box and transported to the Laboratory. For each lot, 1 kg of legume seeds was taken as required, rinsed four times in deionized water to eliminate dust and the insecticide (used for protecting treatment), dried at room temperature and packed in plastic bags and stored at 4 °C until used. All the chemicals used were analytical grade.

2.2. Starch extraction

Starches were extracted from legume seeds by the modified method described by Sathe et al. (1984). In the procedure of starch extraction, 0.5 kg seeds were washed abundantly with tap water, soaked for 12 h, manually dehulled and ground to obtain a paste. Water (5 L) was then added to the paste and mixed manually. The mixture was kept for 12 h during which the starch settled. The precipitated was then suspended in 1 L NaCl 1% and gently mixed. The mixture was kept for 12 h, centrifuged (3800 g, 20 min, 20 °C). The precipitate was collected, washed twice with distilled water, suspended in 1 L NaOH 0.03 M, gently mixed and kept at 4 °C for 12 h. The starch solution was again centrifuged at 3800 g for 20 min. The precipitate was washed with distilled water through a sieve mesh 125 µm to eliminate fibers and the obtained solution was centrifuged (3800 g, 20 min, 20 °C). The resulting precipitate was dispersed on a tray and dried in an air convection oven (Imperial V) at 45 °C for 45 min, grounded into powder in a mortar, packaged in polyethylene bag and stored at 4 °C until used. The total starch content was measured according to AACC Method 76-13.

2.3. Amylose content

Amylose content of the isolated starches was determined by iodine binding method as previously showed by Williams et al. (1970). The starch sample (20 mg/on dry matter) was dispersed in 10 ml potassium hydroxide (0.5 N), vortexes for 5 min, and the volume was made up to 100 ml using distilled water. 10 ml of aliquot was taken and added 5 ml hydrochloric acid (0.1 N) and 0.5 ml of iodine reagent (20 g potassium iodide and 2 g resublimed iodine were dissolved in 100 ml distilled water, further 10 ml of this solution diluted to 100 ml was used as iodine reagent) were added and made up to 50 ml. Absorbance was measured at 625 nm (Shimadzu UV-1601 Spectrophotometer, Japan). Amylose content (%) was determined from a standard curve developed using amylose and amylopectin blends.

2.4. Light transmittance

This was determined using the modified method of Perera and Hoover (1999). A 1% aqueous suspension of starch was heated in a water bath at 90°C for 1h with constant stirring. The suspension was cooled for 1h at 30°C. The samples were stored for 24 hours at 4°C in a refrigerator and turbidity was determined by measuring the absorbance at 640nm against a water blank with Shimadzu UV-1601 Spectrophotometer, Japan). The absorbance measured was interpreted as presented in Equation 1:

$$\frac{\text{Transmittance Percentage}}{100} = 10^{-\text{Absorbance}} \quad (1)$$

2.5. Starch gel strength

The resistance to penetration of the gel was determined with a Model 4500 R LFRA Texturo Analyzer testing machine equipped with a data acquisition and processing station. The SON load cell was used. The gels prepare precedently and store 24h at 4°C was allowed to stay at room temperature during 2h. The gels were transferred within the aluminum molds were placed on the compression table. The cross-head of the machine, fitted with the load cell and a cylindrical probe (5 mm diameter), was driven down so as to just touch the gel surface. The probe was then driven at a constant speed (1 mm/min) into the gel for a distance of 7 mm. The load at 1 mm compression was termed firmness (maximum penetration force). The resultant readings, gel strength (positive area under the force-time curve) were in units of load grams. The results are the means of four replicates.

2.6. Freeze-thaw stability of starch

Stability under refrigeration and freezing conditions was evaluated using a modified version of Eliasson and Kim (1992) method. All starch gels were prepared using the same procedure described before. 100mg of each starch sample gel was transferred to a respective 25 mL centrifuge tube. After that, the tubes were frozen at -20°C for 24 h in a freezer and thawed in a constant temperature incubator at 30°C for 2 h. The tubes were centrifuged at 8000g for 10 min in a J2- HS centrifuge (Beckman Instruments, Inc. CA, USA) and measurements taken of water separation from the starch gels. The above Freeze-Thaw Cycle (FTC) was repeated 1, 2, 3, 4 and 5 days. Syneresis was calculated from the formula of Equation 2:

$$\text{syneresis} = \frac{\text{Weight of release liquid}}{\textit{Weight of starch paste}} \times 100 \qquad (2)$$

2.7. Morphology of starch granule

Scanning electron microscopy (SEM) studied the surface morphology of the starch samples. SEM micrographs was recorded with a Jeol-JSM 6601ALV (made in Japan). Starch samples were applied on an aluminum stub using double sided adhesive tape, and the starch was coated with gold (using JFC 1600 gold coater). The micrographs were obtained with an accelerating potential of 5 kV under high vacuum. In the present study a magnification of 1000X was used for all starch sample.

2.8. Particle size distribution

Particles size distribution was measured with a laser light diffusion apparatus (Mastersizer S, Malvern Instruments Ltd, Malvern, UK) comprising a 5 mW He-Ne laser operating at a wavelength of 632.8 nm with a 300RF lens. Starch was dispersed in ethanol and the sample dispersion unit for small volume was used. The instrument output used volume distribution, with median diameter (D50). This D50 means that the diameter of the particles was smaller for those which had flour volume of 50 mL/100 mL. The derived output data are numerical transformations of diffracted light angles according Mie's theory (Malvern Instruments Ltd, 1990). The dispersibility index (span) of the starch powders was also calculated as shown below in Equation (3) and gives information on the uniformity of the distribution.

$$\text{Span(-)} = \frac{D90 - D10}{D50} \qquad (3)$$

2.9. Thermal properties

The thermal properties of isolated legume starches were analyzed using Differential Scanning Calorimetry (PerkinElmer DSC 4000). Starch samples were weighed in an aluminum pan and distilled water was added with the help of a micro syringe to obtain a starch water suspension

containing 70% water (w/w). The pan was hermetically sealed and allowed to equilibrate for 1h. The instrument was calibrated using indium and empty aluminum pan was used as reference. The sample pans were heated from 40 to 110°C at the rate of 10°C/min the onset of gelatinization (To), the temperature at peak (Tp), the temperature at the end of gelatinization (Tc) and enthalpy of gelatinization (ΔH) were determined.

2.10. Statistical analysis

All experiments were replicated three times for each starch sample. When relevant, a statistical analysis was performed on the treatments through analysis of variance (ANOVA). Principal Components Analysis was performed to evaluate which and how the evaluated factors correlate with the starch studied. Statistica 12.0 (StatSoft) software was used for these analyses.

3. Results and Discussion

3.1. Total starch content (TC) and amylose content (AC)

Starch contents of the isolated legume starches showed no significant difference and ranged from 82.06 g/100g for White bean to 88.40 g/100g for Yellow bean (Table 1). This revealed the high purity of legumes starches extract (Saho Tsaju Nguetcho et al., 2019). It's rather difficult to obtain pure starches from legumes, due to their high protein content. The total amylose content of tested starches was high and varied significantly ($p < 0.05$) from 26.21 g/100g for Yellow bean to 44.85 g/100g for Red spotted bean. High amylose content for a starch means high viscoelasticity and low retrogradation for the gel of the starch. Retrogradation of gel minimized the quality of industrial products while the viscoelasticity enhanced the quality of starch-based products This characteristic of legume starches studied represent therefore a desirable parameter of utilization for industrialists. This result is in accordance with previous reports literature because legume starch usually has high amylose content (Demiate et al., 2016; Ngobese et al., 2017). The difference in amylose content of isolated starches could be influenced by genotypes, soil and climatic conditions (Reddy et al., 2017).

3.2. Light transmittance (LT)

Light transmittance (Table 1) indicates the clarity of cooked starch after 24 hours of storage at 4°C. The value of light transmittance ranged from 2.6 to 81.42% for different native legume starch (Table 2). Red bean, Yellow bean and Green bean have lowest values of clarity and also content lowest amount of amylose content. Similar result was obtained by Zhang et al. (2017) for 4 legume starch. In fact, we denoted any significant correlation between light transmittance and total amylose content. Black bean and Red spotted bean obtained Light transmittance values of 11.54% and 11.04% respectively with no significant difference ($p < 0.05$). The highest transparency value which corresponds to the lowest opacity value was observed in Bambara Groundnut native starch. Cowpea and White bean also had high values of clarity. Light transmittance provides the information on the behavior of starch paste when the light passes through it and depends on granule size, amylose content, granule morphology, and the level of swollen and non-swollen granule remnants (Wani et al., 2016; Yu et al., 2018). The presence of phosphate monoester in legume starch molecule can influence the transparency of gel starch (Jane et al., 1999).

3.3. Gel texture

For the eight native legume starch studied (Table 1), Bambara Groundnut formed a most firm and strong gel (1372.75 g and 140.02N), followed by Cowpea starch gel (1057.75 g and 107.89N). Some modified starches like hydroxypropyl starch and acid treated starch are usually add in food as stabilizers and thickeners; Bambara Groundnut and Cowpea gels can replace that modified starches with these fonctionalities and therefore improve the quality of novels food. The six common bean starch gels firmness and strength values range from (326.75 g to 562.25g) and (33.32 N to 57.34N) respectively. This result is in adequacy with result obtained in the literature for bean gels (Ngobese et al., 2017). Common bean cold gels in this study were significantly ($p < 0.05$) higher than those of maize, rice and potato starch which are the most commercially used starches. We observed a high positive correlation between texture properties and the clarity of gel ($r=0.86$; $p < 0.05$). This confirm the possibility of adding Bambara Groundnut and Cowpea starch gel in frozen food and enhance their color (high transmittance), test (without any chemical modification) and texture (high firmness and gel strength).

3.4. Freeze-thaw stability

The freeze-thaw factor reflects the ability of gel starch to resist rough and successive physical changes of temperature. Free water in starch gel forms ice crystals during freezing; during tawing, that water is expelled from the starch gel network induising the phase separation and the gel dehydration (Ye et al., 2016). Successive

Table 1. Total starch content, amylose content, light transmittance, gel strength and firmness legume starches.

Variety	Total Starch Content (%)	Amylose Content (%)	Transmittance (%)	Gel Strength (g)	Firmness (N)
Cowpea	84,43±1,69a	32,83±0,65b	45,79±6,72e	1057,75±21,15d	107,89±3,88e
Bambara Groundnut	83,3±1,66a	30,48±0,60a	81,42±9,07f	1372,75±27,45e	140,02±3,18f
Red bean	82,87±1,66a	28,16±0,56a	5,68±2,28b	440,50±8,81b	44,93±6,36b
White bean	82,06±1,64a	33,07±0,66b	36,63±6,01d	394,50±7,89b	40,23±4,94b
Green bean	82,44±1,65a	29,76±0,59a	2,6±0,48a	418,75±8,37b	42,71±5,30b
Black bean	83,78±1,67a	35,96±0,71b	11,54±3,24c	326,75±6,53a	33,32±4,59a
Red Spotted bean	85,21±1,70a	44,85±0,89c	11,04±2,31c	562,25±11,24c	57,34±3,88c
Yellow bean	88,4±1,77b	26,21±0,52a	3,78±1,12a	546,00±10,92c	55,69±5,65c

Mean ± standard, n=3. Means with the same letters in a column do not differ significantly (P<0.05).

Table 2. Particle size distribution.

Variety	D10 (μm)	D50 (μm)	D90 (μm)	Span (-)
Cowpea	11,48±0,23^{d}	20,54±0,14^{d}	91,90±6,56^{c}	3,91±0,31^{a}
Green Bean	16,51±0,43^{b}	26,99±0,25^{c}	73,43±32,29^{d}	2,10±1,17^{b}
Black Bean	14,62±0,95^{c}	25,35±0,12^{c}	45,89±5,55^{e}	1,23±0,26^{c}
White Bean	18,3±0,18^{a}	28,38±0,21^{b}	50,95±1,26^{e}	1,15±0,05^{c}
Red Bean	16,49±0,11^{b}	24,97±0,13^{c}	34,87±0,17^{f}	0,73±0,00^{d}
Yellow Bean	17,11±0,77^{a}	26,32±0,26^{c}	39,03±2,57^{f}	0,83±0,06^{d}
Red Spotted Bean	15,06±0,01^{b}	29,82±0,20^{b}	123,86±3,91^{b}	3,65±0,10^{a}
Bambara Groundnut	18,36±0,04^{a}	37,06±0,23^{a}	155,08±7,40^{a}	3,69±0,18^{a}

Mean ± standard, n=3. Means with the same letters in a column do not differ significantly (P<0.05).

freeze–thaw cycle (FTC) accentuated the phase separation and water released from starch gel. We observed that gels from 8 native legume starches were all stable after 3 FTC (Figure 1). However, for the first FTC, the percentage of total water loss ranging from 40% for Cowpea gel to 58% for Black bean and Bambara Groundnut gels. At the fifth FTC, syneresis ranged from 47% to 60% for Cowpea and Black bean starch paste respectively. Tang et al. (2018) obtained 63.3% of syneresis for Wheat starch after adding 8% of exopolysaccharide to reduce the release of water. Cameroon legume starch could therefore withstand the FTC without modification and be more suitable for frozen starch-based food products. In general, syneresis increases with the number of FTC due to the extent of the separation phase, accelerated by the retrogradation of the starch (Srichuwong et al., 2012). There was no increase of amount of water release from legume starch after the third FTC. The high stability of legume could be improved by their high amount of amylose content and their high water absorption capacity (Saho Tsaju Nguetcho et al., 2019). Indeed, the syneresis of starch gels is similar to the syneresis of waxy corn of 53.9% (Srichuwong et al., 2012). Amylopectin or waxy starch is mainly used in food products, but also in the textile, adhesives, corrugated board and paper industries. Since the native starches of our local legumes have this sought-after quality for chilled and frozen products, they can mainly replace these modified starches in the food and pharmacological industries.

3.5. Morphology of starch granule

Scanning electron microscopy (SEM) images of isolated starch granules from each legume starch extract showed no evidence of starch damage, devoid of broken granules, no visible cracks or indentations on the surfaces of granules (Figure 1). The surfaces appeared to be smooth and showed no evidence of fissures when viewed under the SEM. All starch extract showed granules of spherical, oval or elliptical-shapes, kidney-shaped, dome-shaped with smooth surfaces. Generally, the smaller granules were spherical in shape compared to their larger counterparts, which were oblong to elliptical in shapes (Figure 2). Similar granular morphologies of isolated bean starch have been reported previously (Reddy et al., 2017; Ma et al., 2017; Hoover and Sosulski, 1990; Sathe et al., 1984). Generally, legume bean starch granules have morphologies nearly similar, but very different from other starches such as starches of tapioca and banana (Ratnayaye et al., 2002).

Granule clustering was evident in Cowpea and Red bean starch, similar to that reported by Ashogbon and Akintayo (2013) for Cowpea also by Chavan et al.; (1999) for beach pea, green pea and grass pea. In contrast, there was no clustering tendency in other granules of legume starch studies. Martins et al. (2020) observed clustering of Taro starch granules after ultrasonic modification and Zhong et al. (2022) observed aggregation of rice starch granule after adding citric acid at 40%. Native Cowpea starch can therefore be requiring in cooking food needing light viscosity.

3.6. Granule size distribution

Granule size distribution of starches from legume is shown in Figure 3. The starch frequency curves of starch granule size are relatively similar and show a bimodal distribution. The bell shape of all the curves reflects a homogeneous distribution of the granules of the starches analyzed. This distribution of legume starches reflects the fact that the starches studied are made up of two large classes of granule sizes. In most cases, the starch granules exhibit a major small particle size distribution and a minor secondary peak form of bigger particles. These observations were similar to those reported for some varieties of legume starches isolated (Reddy et al., 2017; Ma et al., 2017; Ratnayaye et al., 2002).

Cowpea has the smallest class of granule size ranging from 11.48 to 20.54µm and Bambara Groundnut has the largest class from 18.36 to 37.06µm. Small granules are very useful in industries. In fact, studies have shown that a starch rich in small granules constitutes a good fat substitute. It is also utilized in the manufacture of biodegradable plastics and aerosols as well as in the cosmetics, pharmaceutical and textile industries (Moorthy, 2002).

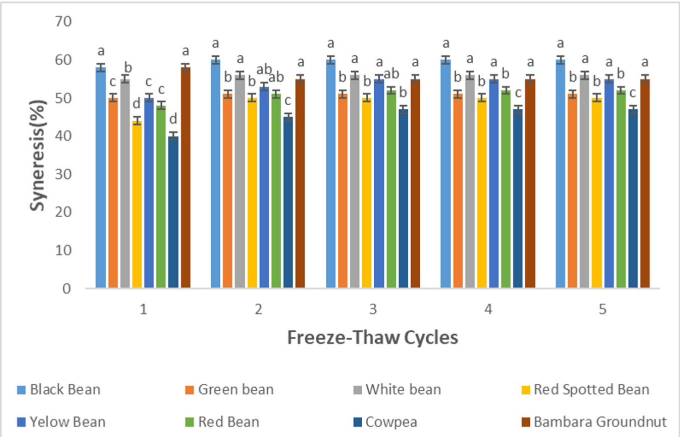

Figure 1. Syneresis of eight Cameroon legume starch gels involving five freeze-thaw cycles. Bars with the same letters do not differ significantly (P<0.05).

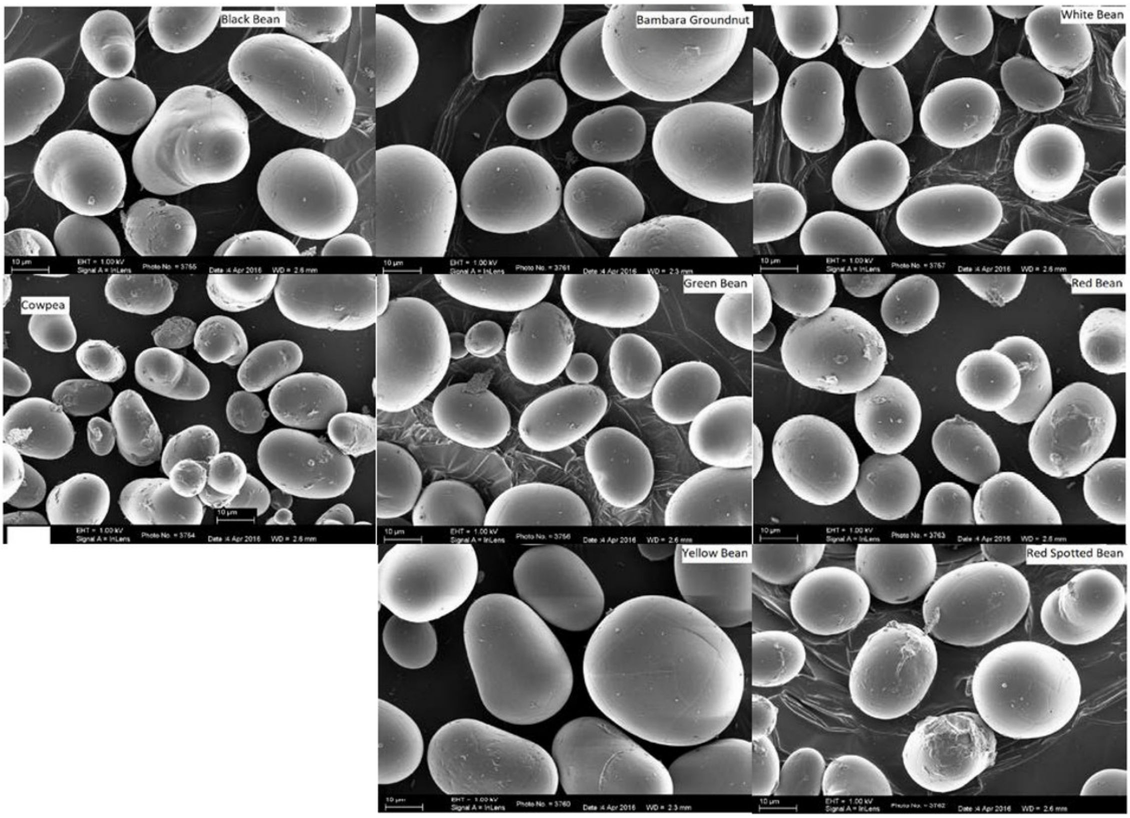

Figure 2 . Scanning electron micrographs of eight Cameroonians native legume starches.

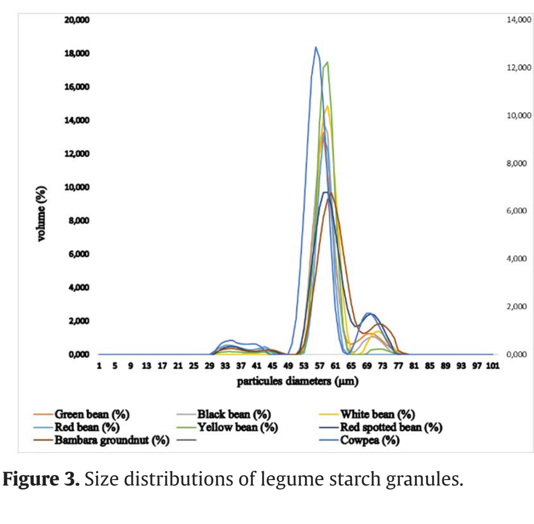

Figure 3. Size distributions of legume starch granules.

In the genus Phaseolus studied, White bean has the largest starch sizes of 18.3 to 28.38µm and Black bean the smallest starch sizes ranging from 14.62 to 25.35µm. These results are close to those obtained by Wang et al. (2019) for the starch of five varieties of Phaseolus vulgaris with peak granule size close to 28 µm and for the starch of four legumes (lentils, peas chick and two varieties of Faba bean) with peaks between 19.6 and 23.9µm. Additionally, Red bean and Yellow bean have low Span values which denote a very small difference in the sizes of their starch granules; while Bambara Groundnut, Cowpea and Red spotted bean have high Spans characterizing a more varied distribution in size of the starch granules. The size and shape of the starch granules are important characteristics, which prominently influence their paste viscosity, thermal transitions and functional properties (Rasper, 1971).

3.7. Thermal properties

The gelatinization thermograms and histograms of native starches from different legume varieties are shown in Figure 4, and the thermal transition temperatures, onset temperature (To), peak temperature (Tp), conclusion temperature (Tc), gelatinization temperature range (ΔTr) and gelatinization enthalpy (ΔH) on a dry starch basis are presented in Table 2. The DSC endothermic peak is linked to starch gelatinization which can reflect the loss of ordered structures like double helices of crystallites of starch granules (Cooke and Gidley, (1992)). The gelatinization transition parameters and gelatinization enthalpy of starches from various legume varieties differed significantly as shown in Table 3. Gelatinization temperature is a reflection of the perfectness of starch crystallites (Tester and Morrison, 1990). The To of legume starches ranged from 68.49 to 74.83°C, Tp in the range of 73.00 to 81.00°C,

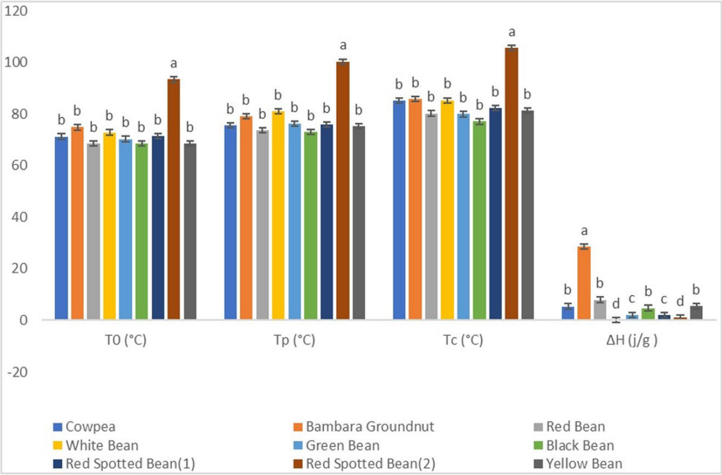

Figure 4. Differential scanning calorimeter graph and histogram of thermal parameters. Bars with the same letters do not differ significantly (P<0.05).

Table 3. Thermal properties of the legume starches.

Variety	T_0 (°C)	T_p (°C)	T_c (°C)	ΔT_r (°C)	ΔH(J/g)
Cowpea	71,25±1,42a	75,67±1,51a	85,25±1,71a	14,00±0,4a	9,11±0,12c
Bambara Groundnut	74,83±1,49b	79,17±1,58b	85,79±1,71a	10,96 ±0,4c	14,68±0,23a
Red Bean	68,49±1,37a	73,67±1,47a	80,21±1,61b	11,72±0,2b	11,31±0,17b
White Bean	72,85±1,46a,b	81,00±1,62b	85,31±1,71a	12,46±0,3b	3,38±0,07f
Green Bean	70,32±1,46a	76,17±1,52a	79,94±1,60b	9,62±0,1c	7,14±0,08d
Black Bean	68,58±1,37a	73,00±1,46a	77,05±1,54b	8,47±0,2d	4,76±0,15e
Red Spotted Bean	71,49±1,43a	76,00±1,52a	82,22±1,64a	10,73±0,2c	6,52±0,08d
Yellow Bean	68,59±1,37a	75,17±1,50a	81,31±1,63a	12,72±0,3b	9,16±0,13c

Mean ± standard, n=3. Means with the same letters in a column do not differ significantly (P<0.05). T_0, onset gelatinization temperature; Tp, peak gelatinization temperature; Tc, conclusion gelatinization temperature; ΔT_r, gelatinization range (Tc _ To); △H, gelatinization enthalpy.

and Tc in the range of 77.05 to 85.79°C, and the Black Bean starch showed the lowest Tp and Tc values among all legume starches studies (Table 3). The lower gelatinization transition temperature of Black Bean starch is indicative of its lower degree of stability. This result concorded with the fact that Bambara Groundnut have the highest granule size.

The legume starch exibited one endothermic DSC micrograph except the Red Spotted Bean starch which shown 2endotherms. The second endotherm determined the dissociation of the fat-amylose complex. This phemomenon is known and researched in food industy.

The DSC curves of Bambara Groundnut starch exhibited wider peaks than those from other seven legume starch (Figure 5), which suggested its broader range of gelatinization temperature (ΔTr) as shown in Table 3. The ΔTr is related to the heterogeneity of the starch granule sizes and the distribution of amylose and amylopectin, which produce a semi-crystalline arrangement inside the granule (Agama-Acevedo et al., 2014). The lower ΔTr value of Black Bean starch may be attributed to the higher homogeneity of crystallites within its smallest starch granules. We obtained a positive high significant correlation r= 0.79 (p≤0.05) between To and D90 (90% of starch granule size). The highest ΔH for Bambara Groundnut starch, whereas the lower were for Red Spotted Bean, White Bean and Black Bean starches. The enthalpy revealed the crystallinity degree and the aspect of molecular order within the granule during gelatinization (Cooke and Gidley, 1992). The lower ΔH indicates a lower degree of organization and a lower stability of the crystals (Chiotelli and Meste, 2002). Bambara Groundnut starch has high gelatinization temperature ranges and highest gelatinization enthalpy than other starch samples and required more energy for gelatinization (Table 2).

3.8. Principal component analysis

The PCA results (Figure 5) showed that firmness clarity and different temparatures of gelatinisation are the variable that presented a higher impact on the gel quality. In addition, it was demonstrated that the particle size (D90) was positively correlated (r = 0.79) with the onset temparature. In the other hand, the FTC parameters were

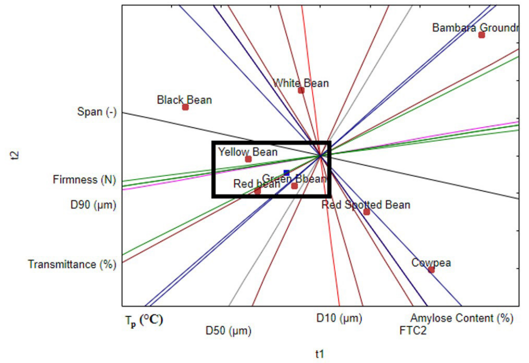

Figure 5. Principal component analysis represented on two principal components.

positively correlated to the small granule size (D10) but negatively impacted the gel quality. Regarding the amylose content and size distribution (Span), they were not strongly represented in the biplot scatter (t1xt2). Therefore, their influence was not evidenced.

4. Conclusion

Structural, morphological and functionnal properties of eight cameroonian legume starches were investigated. High variability among various legume starches properties was found. For instance, Bambara Groundnut starch exibited biggest size granule, strong gel, high temperature of gelatinization and highest enthalpy of gelatinisation with stability during FTC. Bambara Groundnut cameroonians native starch is therefore a best substitute for starches used in food, pharmaceutical and paper industries like binder. Moreover, Cowpea starch possessed the same quality with low syneresis and without a big enthalpy of gelatinisation. Native Cowpea starch is good for Frozen starchy food thickener. Red Spotted Bean with the highest amount of amylose, two endotherms of gelatinisation, big granule size is better for gelification during cooking sauces. The phaseolus Vulgaris starch studied showed slight difference among their charateristics and could be added with insurance in multiple novel food. This research would be interesting to selecting legume starch for specific used, to enhance the scientific and industrial knowledge on Cameroonians' legume starch and finally to reduce the use of chemically modified starches.

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