Physicochemical Properties of Starches from Three Nigerian Commercial Varieties of Cowpea (Vigna Unguiculata) in Comparison with Three Hybrid Types

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ABSTRACT--- Starch isolated from three Nigerian commercial cowpea (Vigna unguiculata) varieties: Drum, Oloyin (sugary), Sokoto, and three hybrid types: ART 98-12, Ife BPC and Ife brown were characterized in terms of x-ray diffraction, electron microscopy, chemical composition, amylose content, swelling power and amylose leaching, RVA paste characteristics, paste clarity and freeze-thawing stability. The starches exhibited A-type diffraction pattern with granule size range 4.71-58.82μm. The starches contained 28.68-35.15% amylose and exhibited high pasting temperatures, restricted swelling and amylose leaching, low paste clarity and poor freeze-thawing stability. The hybrid types showed improved pasting and gel properties for food exploitation.

Keywords--- Cowpea (Vigna unguiculata), starch, physicochemical properties, x-ray diffraction, electron microscopy

1. INTRODUCTION

Cowpea, (Vigna unguiculata L. Walp.) is a tropical grain legume with nutritional importance in developing countries of the subtropics and tropics, especially in Asia, Central and South America and the Sub-Saharan Africa [1]. V. unguiculata plays a vital role in the livelihood of millions of people in West and Central Africa. From its production, rural families derive food, animal feed, and cash income [2]. There is a big market for the sale of cowpea grain and fodder in West Africa [3]. There has been intense research on cowpea grains to maximize its application. Studies have shown it is rich in protein (19.6-25%) and carbohydrate (62.05-65.52%) [4, 5, 6]. Henshaw [7] has evaluated the physicochemical properties of twenty-eight varieties of cowpea from Nigeria and USA origins and observed significant variation in their physical characteristics and chemical composition. The starch content of cowpea has been reported to range from 38-50.7% [8, 9].

Starch can be used as a staple food in human and animal diets, but it can also be used as a raw material in industries [10]. Starch applications and functionality have been reported to be governed by botanical source, granule size distribution, amylose/amylpectin ratio, molecular size of the various components and environmental factors [11]. Research findings have also demonstrated differences in properties arising from differences in varieties. Hence the need to screen varieties to see those with improved physicochemical properties. In this study, the properties of starches from three commercial varieties of cowpea sold in Nigerian market were compared with those from three hybrid types developed by the Institute of Agricultural Research and Training, Moor Plantation, Ibadan, to assess their potential for application as food and industrial starches.

2. MATERIALS AND METHODS

2.1 Sources of research materials

Six varieties of bean seeds grown in Nigeria were used for this work. They consist of three commercial varieties marketed as: Drum, Oloyin (sugary) and Sokoto purchased from Bodija Market, Ibadan; the hybrid types: ART 98-12, Ife BPC and Ife brown were obtained from Institute of Agricultural Research and Training, Moor Plantation, Ibadan. Nigeria.
2.2 Extraction of starch from beans
This was done according to the method of Schoch and Maywald [12] with modification by Nwokocha et al. [13]. The cowpeas were handpicked to clean them of broken pieces and contaminants. 200g seeds was weighed and poured inside a clean bowl, steeped in ordinary water 8 h. The seeds were washed to remove the seed coat. The seeds without coat were blended with excess distilled water for 10 min. The resultant slurry was then filtered through muslin cloth and the starch milk was collected. The starch milk was allowed to sediment and the supernatant decanted. The starch was re-slurred several times in excess 0.1% of sodium hydroxide solution until the supernatant was free of colour. The starch was then washed several times with distilled water until litmus paper confirms it’s free of alkali. Starch was then air dried on a plastic tray. The dried starch was weighed and stored in a closed bowl.

2.3 X-ray diffraction
The starch samples were powdered and sieved to particle size less than 0.074 mm; these were later taken in an aluminum alloy grid (35mm × 50mm) on a flat glass plate. Diffraction diagrams of the samples were obtained using the Rigaku D/Max-III C X-ray diffractometer (Rigaku Int. Corp. Tokyo, Japan) at a scanning rate of 2°/min in the 2 to 50°at room temperature with a Cu Kα radiation set at 40kV and 20mA.

2.4 Microscopy
The starch sample was placed under the electron beam of the Scan Electron Microscope (JOEL JSM – 7600F) and the image was created by absorption, contrast and scattering effects of the sample. The surface of the sample was scanned with an electron beam, and the image was created by secondary electrons emitted from the surface observed. The starch powder on a microscope slide was viewed with a light microscope (objective magnification X40) (Meiji Techno, Japan). The computer managed video was analyzed and the granule properties determined.

2.5 Proximate analysis
Moisture, protein, lipid and ash were determined according to AOAC [14]. Moisture was measured at 105°C using moisture analyzer (AND MX-50, Japan) until it gives a signal indicating the moisture content. Nitrogen was determined by Kjeldahl method and protein obtained as %N × 6.25. Lipid was determined by extraction with hexane using soxhlet apparatus (Thermo Scientific, 10931360 UK) and recovering the lipid by rotorevaporation (Rotavapor B.U.C.H.I R-210, Switzerland). The weight of lipid was expressed as a weight percent of dry starch. Sample from moisture determination was used for ash determination. These were heated in pre-weighed crucibles at 550°C in a furnace (Carbolite type AAF 11/18, England) until completely ashed, for 12 h. The ash content was calculated as percentage weight of ash per dry weight of starch.

2.6 Amylose content
This was done according to the method of Stevenson et al. [15]. 0.1g of sample was placed in centrifuge tube and 1mL of 95% ethanol was added, then 9 mL of 1 M NaOH was added. Solution was shaken vigorously and heated in a boiling water bath for 10 min to get the starch dissolved. It was removed and allowed to cool.
1 mL of starch solution was taken and made up to 10 mL with distilled water. 0.5 mL was then taken from the new solution and placed in 2 different tubes. 0.1 mL of acetic acid was added then 0.2 mL of iodine was also added. 9.2 mL of distilled water was added and solution is left at 25°C for 20 min and shaken again. Percentage of light absorbance was determined at 620 nm against blank using Spectrumlab 22 spectrophotometer and used to calculate amylose as shown in Eq. 1.

\[
\text{Amylose} = \frac{\text{absorbance of sample} \times 0.02 \times 2500}{\text{absorbance of standard}} \quad \text{(g/100g)} \quad \text{(Eq. 1)}
\]

2.7 Paste characteristics
2.7.1 Swelling Power and Amylose Leaching
This was done according to the method of Nwokocha et al., [13]. The swelling power was determined on starch paste by dispersing 0.1g of starch sample in 10mL of distilled water and heated in water bath (HH-S6 Digital Thermostatic Water Bath Gallenkemp, England) for 30 min at temperatures 50, 60, 70, 80, 90°C. The samples were left agitated throughout the heating period to maintain starch suspension. The samples were centrifuged (Centrifuge Model 80-2, Lenfield Medical, England) at 1500 rpm for 10 min. The supernatant was carefully drawn up. The weight of paste was determined and used to calculate swelling power as weight of paste divided by original weight of dry starch.
5 mL of supernatant was then transferred into 100 mL standard flask followed by the addition of acetic acid (1mL) and iodine (2mL) and the volume made up to mark with distilled water. This was shaken and percentage of light absorbance was determined at 620 nm against blank using Spectrumlab 22 spectrophotometer.
2.7.2 Pasting properties
The pasting properties were obtained on a Rapid Visco Analyzer (RVA-S4, Newport Scientific, NSW, Australia). 3.0 g of sample was weighed and 25 mL of water was dispersed into canister. The paddle was placed inside the canister; this was placed centrally onto the paddle coupling and then inserted into the RVA machine. The measurement cycle was initiated by pressing the motor tower of the instrument. The profile can be seen as it is running on the monitor of a computer connected to the instrument. The 13 min profile was used, the time-temperature regime used was: Idle temperature 50°C for 1 min, heated from 50°C to 90°C in 3 min 45 s, then held at 95°C for 2 min 30 s the sample was subsequently cooled to 50°C over a 3 min 45s period followed by a period of 2 min where the temperature was controlled at 50°C.

2.7.3 Paste clarity
This was done according to the method of Singhal and Kulkarni [16]. Pastes (1%) were produced when starch (0.1g) was suspended in 10mL distilled water in screw cap tubes and heated in water bath at 100°C for 30 min. The tubes were thoroughly shaken every 2 min to maintain starch suspension, after cooling to room temperature (5 min), the percentage of transmittance at 650nm was determined against water as blank in a Spectrumlab 22 spectrophotometer.

2.7.4 Freeze-thawing stability
This was done according to the method of Singhal and Kulkarni [16]. The freeze-thaw stability was determined on starch paste by dispersing 0.3 g of starch in 10mL of distilled water in weighed centrifuge tubes, heated in water bath (HH-S6 Digital Water Thermostatic Water Bath Gallenkemp, England) at 95°C, with constant mixing until pasting occurred, for 30 min. The starch was then stored at 4°C for 18 h in a freezer and thawed at 25°C for 3 h and then centrifuged (Model 80-2 Lenfield Medical, England) at 2500 rpm for 10 min and weight of exudates determined over a 3-day period. Freeze-thaw stability was calculated as percentage weight of exudates per weight of paste.

2.8 Statistical analysis
Statistical analysis was done using ANOVA while post hoc test was conducted with Tukey to show significant differences.

3. RESULTS AND DISCUSSION

3.1 Microscopy
The scanning electron micrographs of the beans starches are shown in Figure 1. The starch displayed wide range of granule sizes, while starches of Drum, Sokoto and ART 98-12 may be described as being unimodal, those from Oloyin and IFE BPC and IFE brown showed clear bimodal granules sizes, being a mixture of large and small granules. The granules were spherical to oval in shape with most granules of the order 16.47 - 47.06 µm while the small granules in the bimodal starches were of the order of 4.71 µm. Granule sizes of 7.57- 20.56µm have been reported for Lablab bean starch [13] while Lawal [17] reported granule diameters of 7-40µm for pigeon pea starch. Soybean starch consists of very small granules of 0.7-4µm [15]. The relationship between granule size and starch physicochemical properties has generated great interest in recent times. Smaller granules have been reported to show greater swelling, enzyme susceptibility and faster retrogradation compared with larger ones [18, 19].
Figure 1. Granule micrographs of cowpea starches: A). Drum; B). Oloyin; C). Sokoto; D). ART 98-12; E). Ife BPC; F). Ife brown
3.2 X-ray diffraction
Native starch is believed to display three characteristic X-ray diffraction patterns, A-type, B-type and C-type. An A-type diffraction pattern is seen in many cereal starches and has specific peaks at 15, 17, 18 and 23 while a B-type is seen mostly in tuber starches is associated with peaks at around 5.8, 15, 17 and two small peaks at 23 and 24. However, a C-type X-ray diffraction pattern which is common in pea starches is a mixture of A- and B-type patterns [13]. The six varieties of beans exhibited an A-type diffraction pattern (Figure 2). Also all the starches showed a peak in the XRD at 20° 2θ attributed to amylose-lipid complexation. A C-type diffraction pattern has been reported for some other bean starches: mung bean [20, 21], soybean [15], pigeon pea [17] and lablab seeds [13].

Figure 2: X-ray diffraction of cowpea (Vigna unguiculata) starches
3.3 Proximate composition
Table 1 shows the chemical composition of the starch extracts from cowpea seeds. The moisture and protein contents showed significant difference (P<0.05) among the starch extracts. The moisture content was generally higher than (7.49-8.88%) reported for Phaseolus vulgaris L. [22]. Lipid (0.30-0.70%) and ash (0.33-0.50%) contents showed non-significant difference (P>0.05) among the cowpea starch extracts and values were similar to that of Phaseolus vulgaris L. [22]. The chemical content is a good means of expressing the purity of the starch extracts which shows that lower contents of other components (ash, fat, protein) are highly desirable [23, 24]. It was reported that high contents of other components, especially fat and protein, influence the swelling power and pasting properties of starches [12]. The moisture content is related to the humidity of the environment of storage of the starch and illustrates its shelf life, the lower the moisture content the longer the shelf life.

3.4 Amylose content
Table 1 shows the Amylose content for cowpea starches. ART 98-12 contained the highest amylose (35.15%) while IFE BPC had the least (28.68%). There was significant difference (P<0.05) in amylose content with the cowpea starch varieties. The amylose content of the starches is in the range of values (20.88 - 48.72%) reported for twenty-two stains of cowpea [25] but higher than 22.06-26.53% reported for white and brown cowpea starches [9]. Similar high amylose contents have been reported for other bean starches: 38% for black bean starch [26], 31.0% for D. lablab starch [27]. It has been reported that amylose content is important in controlling the physicochemical properties of starch such as enzyme digestibility, gelatinization temperature, swelling power, viscosity peak, breakdown and setback [28].

Table 1: Composition of the Cowpea (Vigna unguiculata) Starches

<table>
<thead>
<tr>
<th>Cowpea variety</th>
<th>Moisture</th>
<th>Protein</th>
<th>Lipid</th>
<th>Ash</th>
<th>Amylose</th>
</tr>
</thead>
<tbody>
<tr>
<td>Drum</td>
<td>15.89± 0.40a</td>
<td>0.87± 0.02a</td>
<td>0.50±0.14a</td>
<td>0.50±0.24a</td>
<td>29.78±1.37a</td>
</tr>
<tr>
<td>Oloyin</td>
<td>15.57 ±0.17a</td>
<td>0.95±0.01b</td>
<td>0.30±0.14a</td>
<td>0.33±0a</td>
<td>31.32±0.68ab</td>
</tr>
<tr>
<td>Sokoto</td>
<td>16.645±0.53ab</td>
<td>0.90±0.01a</td>
<td>0.50±0.14a</td>
<td>0.50±0a</td>
<td>34.12±0.73bc</td>
</tr>
<tr>
<td>ART 98-12</td>
<td>17.67±0.23b</td>
<td>0.89±0.01a</td>
<td>0.60±0.28a</td>
<td>0.50±0.24a</td>
<td>35.15±0.57c</td>
</tr>
<tr>
<td>Ife BPC</td>
<td>16.095±0.37ab</td>
<td>0.95±0.04b</td>
<td>0.70±0.14a</td>
<td>0.33±0a</td>
<td>28.68±0.04a</td>
</tr>
<tr>
<td>Ife Brown</td>
<td>17.74±0.68b</td>
<td>0.90±0.03a</td>
<td>0.70±0.14a</td>
<td>0.33±0a</td>
<td>33.25±0.62bc</td>
</tr>
</tbody>
</table>

All values are expressed in percentage. All values are mean of duplicate determinations (mean ± SD). Different superscripts (a-c) in the same column represent significant difference at p<0.05
3.5 Swelling power and amylose leaching

Table 2 shows the swelling and amylose leaching patterns of the bean starches at temperatures 50-90°C. There was no significant difference in swelling power in Drum and Oloyin varieties until 80°C while significant difference in swelling occurred in Sokoto, ART 98-12, Ife BPC and Ife brown from 70°C. The greatest swelling occurred in ART 98-12 (20.96 g.paste/g.starch) while the least swelling occurred in Ife BPC (6.00 g.paste/g.starch) at 90°C. Similarly significant difference in amylose leaching occurred in the starches at 70°C except in Sokoto variety which showed significant leaching from 80°C (P < 0.05). The least amylose leached at 90°C was observed in Sokoto (15.83%) while the highest was observed in Drum (18.07%).

Table 2: Swelling power and amylose leaching of the cowpea (Vigna unguiculata) variety starches

<table>
<thead>
<tr>
<th>Temp °C</th>
<th>Drum</th>
<th>Oloyin</th>
<th>Sokoto</th>
<th>ART98-12</th>
<th>Ife BPC</th>
<th>Ife Brown</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Swelling power (g. paste/g. starch)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>50</td>
<td>2.13±0.16a</td>
<td>2.58±0.12a</td>
<td>2.28±0.28a</td>
<td>2.09±0.11a</td>
<td>2.20±0.10a</td>
<td>2.14±0.001a</td>
</tr>
<tr>
<td>60</td>
<td>2.11±0.06a</td>
<td>2.66±0.06a</td>
<td>2.32±0.30a</td>
<td>2.26±0.12a</td>
<td>2.47±0.18a</td>
<td>2.50±0.18a</td>
</tr>
<tr>
<td>70</td>
<td>2.85±0.23a</td>
<td>3.01±0.19a</td>
<td>2.97±0.09b</td>
<td>2.86±0.04b</td>
<td>3.57±0.29b</td>
<td>3.67±0.12b</td>
</tr>
<tr>
<td>80</td>
<td>12.07±0.13b</td>
<td>13.66±0.28b</td>
<td>12.44±0.27c</td>
<td>14.93±0.35c</td>
<td>13.69±0.77c</td>
<td>17.25±0.07c</td>
</tr>
<tr>
<td>90</td>
<td>18.90±0.38c</td>
<td>19.15±0.02c</td>
<td>16.82±0.50d</td>
<td>20.96±0.29d</td>
<td>16.00±1.34d</td>
<td>18.20±1.20c</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Temp °C</th>
<th>Amylose leached (%)</th>
<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>50</td>
<td>0.14±0.06a</td>
<td>1.28±0.11a</td>
<td>0.24±0.06a</td>
<td>0.58±0.12a</td>
<td>0.72±0.08a</td>
<td>0.58±0.01a</td>
</tr>
<tr>
<td>60</td>
<td>0.15±0.07a</td>
<td>1.17±0.09a</td>
<td>0.34±0.06a</td>
<td>0.43±0.08a</td>
<td>0.73±0.10a</td>
<td>0.48±0.01a</td>
</tr>
<tr>
<td>70</td>
<td>0.25±0.07b</td>
<td>1.58±0.04a</td>
<td>1.10±0.05b</td>
<td>2.29±0.02b</td>
<td>5.25±0.06b</td>
<td>10.21±0.25b</td>
</tr>
<tr>
<td>80</td>
<td>17.63±0.05c</td>
<td>15.58±0.07c</td>
<td>13.13±0.04c</td>
<td>15.47±0.09c</td>
<td>15.62±0.26c</td>
<td>15.42±0.11c</td>
</tr>
<tr>
<td>90</td>
<td>18.07±0.12d</td>
<td>17.68±0.08d</td>
<td>15.83±0.19d</td>
<td>17.42±0.14d</td>
<td>17.06±0.09d</td>
<td>17.28±0.09d</td>
</tr>
</tbody>
</table>

All values of swelling power are expressed in g(paste)/g(starch). All values of amylose leaching expressed in g/100g. All values are mean of duplicate determinations (mean ± SD). Superscripts (a-d) represents significant difference at p<0.05.
It can be seen that both swelling power and amylose leaching increased with increase in temperature. This indicates an increase in temperature results in corresponding increase in thermal energy which results in weakening of the intra-granular bonds permitting the absorption of water into the granules resulting in swelling and solubilisation of the low molecular weight starch molecules especially amylose [13]. Difference in swelling and amylose leaching can be attributed to difference in granule architecture, amylose/amylopectin ratio and molecular size and degree of branching of the amylose fraction and amylose-lipid complexes [29].

![Pasting profiles of 12% (w/v) slurry of cowpea starches](image)

**Figure 3. Pasting profiles of 12% (w/v) slurry of cowpea starches**

### 3.6 Pasting characteristics

The pasting profiles of 12% (w/v) and paste characteristics of the cowpea starches are shown in Figure 3 and Table 3, respectively. The pasting temperature- the temperature the heated starch slurry started to swell irreversible ranged from 79.53°C - 82.48°C. There is significant difference (p < 0.05) in pasting temperature. Ife brown starch showed the weakest intra-granular bonds as it started relaxing its bonds first (79.53°C) while Drum and Sokoto showed tightest structure as they relaxed last. These values are higher than 50.2 and 50.3°C reported for brown and white cowpea starches, respectively reported by Ashogbon & Akintayo [9]. The starches attained viscosity peaks during pasting within a time range of 4.37 - 4.7 min but there was no statistical difference in the peak times. However, the peak viscosities ranged from 4499 cP in Ife brown to 6359 cP in Oloyin and the values were statistically different (p < 0.05). The peak viscosity indicates the maximum swelling capacity of the heated starch slurry and therefore is a measure of the thickening ability of the starch. The breakdown ranged from 1906 - 3152.5 cP. The starch pastes of Drum, Oloyin and Sokoto showed significantly higher breakdown in viscosity compared with those of ART 98-12, Ife BPC and Ife brown (p < 0.05). Lower breakdown indicates greater ability of the paste to withstand severe processing conditions during food processing.
Table 3: RVA paste characteristics of 12% slurry of cowpea (Vigna unguiculata) starches

<table>
<thead>
<tr>
<th>Paste characteristics</th>
<th>Drum</th>
<th>Oloyin</th>
<th>Sokoto</th>
<th>ART 98-12</th>
<th>Ife BPC</th>
<th>Ife Brown</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pasting temp. (°C)</td>
<td>81.63±</td>
<td>80.88±</td>
<td>82.03±</td>
<td>82.48±</td>
<td>79.9±</td>
<td>79.53±</td>
</tr>
<tr>
<td>Peak time (min)</td>
<td>4.53±</td>
<td>4.37±</td>
<td>4.43±</td>
<td>4.70±</td>
<td>4.67±</td>
<td>4.4±</td>
</tr>
<tr>
<td>Peak viscosity, $V_p$ (cP)</td>
<td>6318.5±</td>
<td>6359.5±</td>
<td>6160.5±</td>
<td>4967±</td>
<td>5668.5±</td>
<td>4499±</td>
</tr>
<tr>
<td>Trough, $V_T$ (cP)</td>
<td>3088.5±</td>
<td>3348±</td>
<td>3008±</td>
<td>3061±</td>
<td>3629±</td>
<td>2259.5±</td>
</tr>
<tr>
<td>Breakdown</td>
<td>3230±</td>
<td>3011.5±</td>
<td>3152.5±</td>
<td>1906±</td>
<td>2039.5±</td>
<td>2239.5±</td>
</tr>
<tr>
<td>$(V_p - V_T)$ (cP)</td>
<td>130.11±</td>
<td>72.83±</td>
<td>3.54±</td>
<td>74.95±</td>
<td>75.66±</td>
<td>36.06±</td>
</tr>
<tr>
<td>Final viscosity, $V_F$ (cP)</td>
<td>4216±</td>
<td>4524.5±</td>
<td>4295±</td>
<td>5213±</td>
<td>5410±</td>
<td>3828±</td>
</tr>
<tr>
<td>Setback</td>
<td>1127.5±</td>
<td>1176.5±</td>
<td>1287±</td>
<td>2152±</td>
<td>1781±</td>
<td>1569.5±</td>
</tr>
<tr>
<td>$(V_F - V_T)$ (cP)</td>
<td>33.23±</td>
<td>41.72±</td>
<td>70.71±</td>
<td>173.95±</td>
<td>66.47±</td>
<td>113.84±</td>
</tr>
</tbody>
</table>

All values are mean of duplicate determinations (mean ± SD). Different superscripts (a-d) in the same row represent significant difference at p<0.05.
The setback of the starch pastes ranged from 1127 – 2152 cP with Drum starch paste having the least while ART 98-12 the highest and there exists significant difference among the varieties (p < 0.05). Setback is a viscosity increase arising from re-association of solubilized amylose with granule fragments. From the RVA paste characteristics, the commercial varieties generally showed higher viscosity peak and higher breakdown compared with the hybrid types. This indicates the commercial variety starches had weaker granule structure, however, the commercial varieties had lower setback than the hybrid types. Setback is related to the gel properties of the starches; starches with high setback values form firm gels. Gel strength is affected by amylose/amylopectin ratio, granule size and botanical source of starch [30].

3.7 Paste clarity
Table 4 shows the light transmittance of the six varieties of starch extracted. The pastes showed significant difference in clarity (p<0.05). The order of clarity was Ife BPC ≈ Oloyin < Sokoto ≈ ART 98-12 < Drum < Ife brown. The starch pastes exhibited low clarity (8.00 – 12.00%).

Table 4: Paste Clarity and Freeze-thaw stability of the Cowpea (Vigna unguiculata) starches

<table>
<thead>
<tr>
<th></th>
<th>Drum</th>
<th>Oloyin</th>
<th>Sokoto</th>
<th>ART98-12</th>
<th>Ife BPC</th>
<th>Ife Brown</th>
</tr>
</thead>
<tbody>
<tr>
<td>Percentage light transmittance of 1% starch paste measured at 650 nm $\sigma$</td>
<td>10.95±0.07$^c$</td>
<td>8.25±0.07$^a$</td>
<td>9.60±0.14$^b$</td>
<td>9.85±0.21$^b$</td>
<td>8.00±0.14$^a$</td>
<td>12.00±0.14$^d$</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Time</th>
<th>Percentage syneresis at 3% starch paste $\tau$</th>
</tr>
</thead>
<tbody>
<tr>
<td>DAY 1</td>
<td>65.06±1.10$^{bc}$ 66.79±0.98$^{cd}$ 51.77±1.08$^{abc}$ 48.61±1.22$^{ab}$ 36.21±1.11$^{a}$ 41.82±1.83$^{ab}$</td>
</tr>
<tr>
<td>DAY 2</td>
<td>70.27±1.22$^{b}$a 72.22±1.48$^{b}$a 71.84±0.43$^{b}$a 74.35±1.30$^{b}$a 49.75±0.86$^{bc}$ 55.30±0.71$^{bc}$</td>
</tr>
<tr>
<td>DAY 3</td>
<td>74.41±1.42$^{bc}$a 80.59±1.21$^{b}$c 85.08±0.71$^{bc}$c 84.83±1.03$^{b}$c 62.94±0.71$^{cd}$ 69.52±1.41$^{ef}$</td>
</tr>
</tbody>
</table>

All values are mean of duplicate determinations (mean ± SD).
$\sigma$ Different superscripts ($^{ab}$) represent significant difference at p<0.05
$\tau$ Different superscripts $^{ac}$ in a column, $^{ad}$ in a row represent significant difference at p<0.05

This can be attributed to the high amylose content of the starch resulting in high re-association of the solubilised amylose (retrogradation) with resultant formation of cloudy dispersion or due to presence of amylose-lipid complexes which reduced the solubilization of the starch particles. Similar dependence of paste clarity on amylose/amylopectin ratio and amylose-lipid complexes have been reported for other starches [30, 31].

3.8 Freeze- thaw stability
The freeze–thaw stability of the starches is presented in Table 4. The starches exhibited high instability to freeze–thaw cycles, with significant difference among the varieties and across the time of storage (P < 0.05). At the first freeze–thaw cycle, Drum and Oloyin gave the highest amount of exudates while Ife BPC gave out the least. The hybrid types showed greater stability than the commercial varieties. After three freeze–thaw cycles, the amount of exudates was Drum.
(74.41%), Oloyin (80.59%), Sokoto (85.08%), ART 98-12 (84.83%), Ife BPC (62.94%), and Ife brown (69.52%). Ife BPC showed the lowest value.

This illustrates a high retrogradation tendency of the starch isolates with that of the commercial varieties even more severe than that of the hybrid types. The general values show that freeze-thawing stability of hybrid types is better than the commercial varieties aside from ART 98-12 which showed high value. There was correlation between the amount of amylose content and free-thaw stability. Sokoto and ART 98-12 with the high amounts of amylose content showed the highest instability or syneresis [30]. Similar high water losses have been reported for chickpea and white bean starches [32].

4. CONCLUSION

The starches from the six varieties of cowpea (Vigna unguiculata) had A-type X-ray diffraction pattern. The starch granules were round to oval in shape with those of Oloyin, Ife BPC and Ife brown being bimodal in size. The starches had high amylose content and exhibited restricted swelling. The high retrogradation of the starches showed that they have potential for application as gelling agents in the food industry.

5. REFERENCES


