Infrared heating under optimized conditions enhanced the pasting and swelling behaviour of cowpea starch

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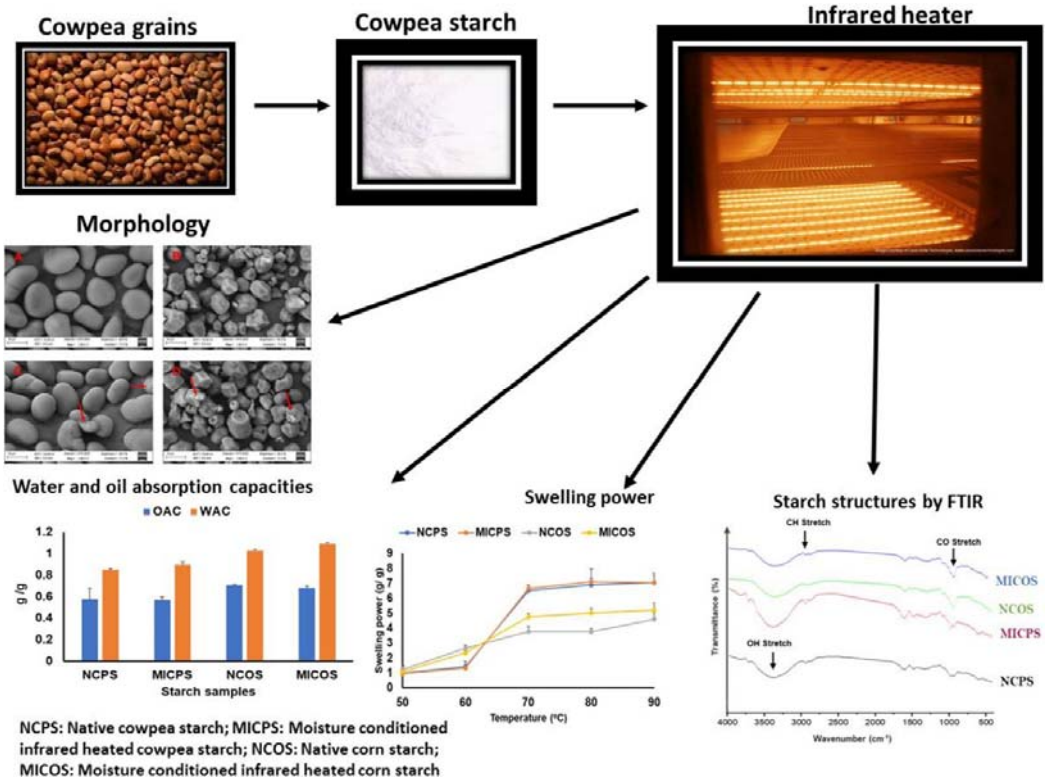
Highlights

•Optimum moisture level & heating time were 46.2 g and 32.9 min, respectively.
•Infrared heated cowpea starch showed higher swelling and pasting properties.
•Cowpea starch was bigger and showed higher swelling power than corn starch.
•Corn starch exhibited higher water absorption capacity and pasting properties.
•Modified cowpea starch had reduced crystallinity & gelatinization temperatures.

Abstract

Native starches are not suitable for industrial use and must be modified for improved functionality. In this study, the effect of moisture preconditioning and infrared heating time on physicochemical properties of cowpea starch was investigated using a two-factor central composite rotatable design. Factors (moisture levels:10–40 g/100 g starch and infrared heating time:10–60 min) with their corresponding α mid-point values resulted in 13 experimental runs. Selected functional and pasting properties were determined as response variables. Starch samples produced under optimized conditions were compared with corn starch and their physicochemical properties determined. Except for pasting temperature, cowpea starch prepared using the optimal conditions (moisture: 46.21 g/100 g starch, dry basis and heating time of 32.88 min) had higher functional and pasting properties compared with the native cowpea starch. Infrared heating significantly reduced the gelatinization temperatures of cowpea starch but did not significantly change that of the corn starch. The crystallinity and double-helical order structure of moisture conditioned cowpea starch also reduced after modification. Cowpea starch showed a bigger granule size, higher swelling power but lower water absorption capacities and pasting properties compared with the control. The infrared heating process is a novel and promising modification method for improving the swelling properties of starch.

Graphical abstract



Keywords: Cowpea starch; Infrared heating; Moisture preconditioning; Thermal properties; Pasting properties

1. Introduction

Cowpea (*Vigna unguiculata*) is a leguminous crop grown in tropical regions of the world and many parts of Africa including Southern Africa. It is the most cultivated legume in Sub-Saharan Africa [1] and has been used to make traditional dishes in various forms [2]. Cowpea has been used in combating protein-energy malnutrition and in improving the nutritional intake of the malnourished [1]. It is an inexpensive source of protein (19–40%) and carbohydrate (50–65%) [3] with the bulk of carbohydrate as starch, varying between 11 and 66% [4], [5], [6].

With the growing demand for alternative starch sources, pulse starches such as those isolated from cowpea can play a significant role in the food industry, especially, because they are rich in resistant starch (9.42–32.14%) [1], [7], [8]. Resistant starch has beneficial physiological effects on humans such as improving the absorption of minerals, preventing colon cancer and supporting the growth of probiotics [9]. Furthermore, cowpea starch is reported to produce strong and firm gels than corn and potato starches, indicating their potentials in confectioneries and as thickeners in food applications [10]. However, the native starch has very limited applications due to the inherent nature of poor resistance to processing conditions which are prevalent in the industry. These limitations include a high tendency towards retrogradation, poor freeze-thaw stability, low process tolerance and gel opacity that prevent its use in food processing [11]. This limitation has prompted researchers and the industry to modify starch using genetic, chemical, enzymatic, physical methods and in some

instances, a combination of these methods to improve starch functionality. Previous studies on cowpea modification have focused on acetylation [12], pyrodextrinization [3], gamma irradiation [13] and autoclaving-cooling cycles [7]. Due to consumers' safety considerations, research efforts have been tailored towards using methods that are safe and environmentally friendly for starch modification. An example of such a method with safety considerations in mind is the use of physical methods such as autoclaving-cooling cycles [7], annealing [14], [15], microwave heating [16], [17] and gamma irradiation [13]. Previous studies on gamma irradiation of cowpea starch reported a significant increase in gelatinization temperature and water absorption [13]. These authors further reported a significant reduction in peak, breakdown, final and setback viscosities with increasing irradiation dose.

The application of physical methods such as infrared heating otherwise known as micronization to modify grain structure and to reduce cooking time has been reported for legumes such as lentil [18], cowpea [6], [19], [20] and Bambara groundnut [18]. Infrared heating is a rapid, high-temperature hydrothermal processing method that uses moisture, temperature and mechanical pressure to achieve starch gelatinization in grains [21]. It is an emerging technology that involves short-time exposure of moisture-conditioned food material to electromagnetic radiation in the wavelength region of about 1.8–3.4 μm [22]. The energy produced is absorbed by the moisture conditioned food materials and this causes vibration and intermolecular frictions among the water molecules and other constituents, resulting in heat generation and a rise in water vapour pressure in the food [19]. This technique has been successfully applied to pulses such as cowpea (*Vigna unguiculata*), kidney bean (*Phaseolus vulgaris*), lentil (*Lens culinaris*) and split peas (*Pisum sativum*) with significant improvement in the cooking characteristics of the various seeds and changes in microstructure and functionality of resulting flours [19].

Ogundele [19] reported a significant reduction in pasting viscosity, due to the presence of denatured protein preventing starch hydration and dispersion during pasting. Starch is an important component of cowpea grains and infrared heating of hydrated cowpea before starch extraction resulted in poor starch functionality, especially at a higher temperature of 170 °C [6]. Thus, it is hypothesized that infrared heating of moisture conditioned cowpea starch may give better functionality than starch isolated after infrared heating of the grain. During infrared heating, moisture levels, heating time and temperature are important variables that may influence the behaviour of the infrared heated samples. The novelty in this study lies in the application of infrared heating as a new method to modify native starch extracted from grains, rather than on structure and flour functionality of cowpea grains or starch extracted from infrared heated grains has reported in an earlier study. Mwangwela et al. [6] studied the effect of different infrared heating temperatures (130, 153 and 170 °C) on the cooking characteristics of cowpea and physicochemical properties of starch isolated from the heated grains. The authors found that although the heating process significantly reduced the cooking time and thus the time required for the cowpea seeds to attain a suitably soft texture, the starch isolated from heated cowpea showed no birefringence indicating the occurrence of gelatinization [6]. Furthermore, the starch isolated from the infrared heated cowpea grains showed higher pasting viscosities compared to starch from cowpea grains that were not heated. Infrared heating has a major advantage of high penetration power and it is highly efficient in achieving high temperatures in a very short time [19]. According to Ogundele [19], several factors such as plate distance, infrared heating power, surface temperature, air velocity, moisture content and size or thickness of the food samples can influence the efficiency of infrared heating.

To the best of our knowledge, no study has been done on the effect of infrared heating on starch isolates, especially from cowpea grains. Hence, this study aims to determine the effects of moisture levels and infrared heating time on structural, functional, pasting and thermal properties of cowpea starch. Corn starch was included as a reference sample since it is one of the main sources of starch for the food industry.

2. Materials and methods

2.1. Materials

Cowpea was obtained from the Agricultural Research Council, Institute for Tropical and Subtropical Crops, Nelspruit, South Africa. The grains were immediately transferred in jute bags to a cold room controlled at 4 °C until the grains were needed for starch extraction. Normal corn starch was obtained from Sigma-Aldrich (St. Louis, MO, USA). All other chemicals and solvents used were laboratory grade.

2.2. Starch isolation

Cowpea starch was extracted as previously reported except that the grains were soaked for 24 h and dehulled manually before milling [7]. Briefly, Dehulled grains were ground in a blender using distilled water (ratio of seed:distilled water = 1:3) into a slurry. The slurry was filtered through muslin cloth and the resulting suspension allowed to settle overnight at 4 °C. The supernatant was drained off. The starch sediment was redissolved in 0.05 M NaOH, centrifuged and neutralized with 2 M HCl to pH 7. Thereafter, the starch sediment was rinsed with distilled water and allowed to settle at 4 °C overnight until the settled starch gave a firm and dense deposit on the bottom. Extracted starch was dried at 50 °C in a hot air oven (D-37530, Thermo Fischer Scientific, Pretoria, South Africa) for 24 h. Dried starch was stored at 4 °C until analysis.

2.3. Infrared heating process

A two-factor central composite rotatable design with 5 centres of rotation ($\alpha = 1.414$) was used to design infrared heating experiments using Minitab 17.0 software (Minitab Inc. State College, PA, USA). Factors and their corresponding levels were moisture level (10–40 g/100 g starch, dry weight basis) and infrared heating time (10–60 mins) with their corresponding α mid-point values (Table 1), bringing about 13 experimental runs. Preliminary studies were done at varying moisture levels and literature was consulted to establish a moisture range and infrared treatment time at a constant infrared heating temperature of 100 °C. Cowpea starch was hydrated to the specified moisture levels and was placed in a laboratory batch infrared heater (MW184, Delphius Technologies, Pretoria, South Africa) at varying time as depicted for each run (Table 1). Infrared heated starches were grounded and the functional and pasting properties were evaluated and the resulting data used as response variables to evaluate the experiment.

Table 1. Experimental conditions for starch moisture conditioned and infrared heat treatment.

Factors	Levels					
	Codes	−α	−1	0	1	α
Moisture level (g)	A	3.78	10	25	40	46.21
Infrared heating time (mins)	B	5.68	10	37.5	60	69.31

2.4. Water absorption and oil absorption capacities

Water absorption capacity was determined by the method described by Oyeyinka [23]. Starch (1 g dry basis) was mixed with 10 mL of distilled water. The mixture was allowed to stand at room temperature for 30 mins and thereafter centrifuged at 4000 ×*g* (5702 R-Eppendorf Centrifuge, Hamburg, Germany) for 30 mins. Water absorption capacity was expressed as a gram of water-bound/g of flour. The same procedure was repeated for oil absorption capacity, except that the water was replaced with soybean oil.

2.5. Swelling power

Swelling power was determined as previously reported [23]. Briefly, a 1% starch suspension in water was heated for 30 mins at 70 °C and subsequently for optimized samples between 50 and 90 °C with constant stirring. The suspension was centrifuged (5702 R-Eppendorf Centrifuge, Hamburg, Germany) at 4000 ×g for 30 mins at 25 °C and the supernatant discarded. Swelling power was obtained by weighing the residue after centrifugation and dividing by the original weight of starch on a dry weight basis.

2.6. Pasting properties

A Rapid Visco-Analyzer (RVA 4500-Perten Instruments, Warriewood, Australia) was used for the determination of pasting properties of the starch samples. Starch (3 g, dry basis) was mixed with 25 g distilled water in aluminum pans. The slurry was mixed with the paddle for at least 2 mins before testing. A programmed heating and cooling cycle were carried out. Samples were held at 50 °C for 1 min, heated to 95 °C for 7.5 mins, kept at 95 °C for 5 mins, cooled back to 50 °C for 7.5 mins and held at 50 °C for 2 mins [24].

2.7. Numerical optimization of moisture and infrared heating conditions

A general model for two-factor central composite rotatable design (Eq. (1)) was adopted in the optimization of moisture and infrared heating conditions and the determination of coefficients of regression.

$$Y = \beta_0 + \beta_1 A + \beta_2 B + \beta_{12} AB + \beta_{11} A^2 + \beta_{22} B^2 \quad (1)$$

where Y is the response, β_1, β_2…β_{22} are coefficients of regression while A and B are experimental factors.

Optimum conditions were obtained by minimization (setback viscosity, breakdown viscosity, trough viscosity, swelling power and oil absorption capacity) and maximization (water absorption capacity, pasting temperature, peak and final viscosities) of responses to achieve the highest desirability factor.

2.8. Analyses of optimized infrared heated starch and controls

2.8.1. Starch composition

Ash (Method 923.03), moisture (Method 934.01), crude protein (Method 990.03), crude fat (Method 920.39) and crude fibre (Method 978.10) contents of the native and modified starch

samples were determined using standard method previously reported [25], while the total starch (Method 76-13.01) was analysed as earlier documented [26].

2.8.2. Functional and pasting properties

The WAC, OAC, swelling power and pasting properties of the optimized samples were determined as described above (2.3 Infrared heating process, 2.4 Water absorption and oil absorption capacities, 2.5 Swelling power). The optimized conditions for the hydrothermal treatment was a moisture content level of 46.21 g and an infrared heating time of 32.88 mins.

2.8.3. X-ray diffraction (XRD)

X-ray diffraction patterns of the starch samples were determined using a diffractometer (Panalytical, Eindhoven, North Brabant, Netherlands) as described by Oyeyinka [23]. Samples were equilibrated at 25 °C and relative humidity of 100%, in a low-temperature incubator (MTIE10, Labcon, Pretoria, South Africa) for 12 h to account for differences in the moisture content of the samples. Equilibrated samples were scanned over a region of 4 to 40 (2θ) ° at a scanning speed of 0.06°/min and operating conditions of 45 kV, 40 mA and CuKα1 (0.154 nm).

2.8.4. Fourier transform infrared spectroscopy (FTIR)

Structural analysis of the starch using a spectrometer (4100-JASCO Spectrometer, Japan) was done according to the method reported previously [23] The spectra were obtained in the transmittance mode with 64 scans from 500 to 4000 cm^{-1}.

2.8.5. Microscopy and amylose content

Starch granule morphology was examined using a scanning electron microscope (EVO 15 HD, CarL Zeiss, Jena, Germany) with an accelerating potential of 4 kV. Briefly, a thin layer of the starch granule was mounted on the aluminum specimen holder with double-sided tape. Starch samples were coated with a thin film of gold for 2 mins with a thickness of about 30 nm [27]. The amylose content of the native and optimized starches were determined using the iodine binding method of Williams [28].

2.8.6. Thermal and pasting properties

The thermal properties of the starch samples were determined at the University of Pretoria, South Africa using a high-pressure differential scanning calorimetry with STARe software (HP DSC827^{e}, Mettler Toledo, Greifensee, Switzerland) as described by [29]. Briefly, about 9–10 mg (db) sample was weighed into a DSC pan and about 27–30 μL of water was added and equilibrated for 24 h at ambient temperature before analysis. Samples were run under pressure (4 MPa) at a heating rate of 10 °C/min from 20 to 120 °C. Thermal properties determined include onset temperature (To), peak temperature (Tp), conclusion temperature (Tc) and enthalpy (ΔH). The pasting properties of the optimized samples were determined as described above (Section 2.5).

2.9. Statistical analysis

Triplicate samples were prepared, and analyses were done in triplicate. Data were analysed using one-way analysis of variance (ANOVA) and means were compared using the Fisher Least Significant Difference (LSD) test ($p \leq 0.05$) using the Statistical Package for the Social Sciences (SPSS) Version 16.0 for Windows (SPSS Inc., Chicago, IL, USA).

3. Results and discussion

3.1. Functional and pasting properties of infrared heated cowpea starch

The different experimental runs for cowpea starch produced under conditions of moisture levels (10–40%) and infrared heating time (10–60 mins) and the functional and pasting properties of the infrared heated starches are presented in Table 2. The ANOVA indicated that moisture levels and time had significant effects ($p \leq 0.05$) on water absorption capacity (WAC) and oil absorption capacity (OAC) (Table 2).

The WAC increased with an increase in moisture levels and infrared heating time. Cowpea starch hydrated at a moisture level of 40 g and infrared heated for 60 mins displayed the highest WAC (0.99 g/g), while the lowest WAC (0.84 g/g) was observed in the cowpea starch hydrated to a moisture level of 10 g but infrared heated for 15 mins. The OAC of the infrared heated cowpea starch generally increased, compared with the control (0.56 g/g). According to Abu [13], starch does not have non-polar sites similar to those found in proteins, hence, the mechanism of oil absorption relies mainly on physical entrapment of oil within the starch structure. Therefore, the increase in WAC and OAC of cowpea starch after moisture preconditioning and infrared heat treatment suggests a possible loosening of the starch structure. The increase in WAC may be due to the increase in amylose content of the starches after modification (Table 3). This seems plausible since amylose content of starches has been suggested to influence their ability to absorb water [23]. Similarly, high WAC has been reported for chick pea [30] and Bambara groundnut starches [23] containing higher amylose content. Furthermore, the increase in WAC may also be partly due to infrared heat-induced changes resulting in the depolymerization of starch to simpler molecules such as dextrins and sugars that have a higher affinity for water absorption. These molecules presumably have more affinity for water absorption than starch and such changes have been reported for microwaved heated [31] and gamma-irradiated legume starches [13].

To: onset gelatinization temperature, Tp: peak gelatinization temperature, Tc: conclusion gelatinization temperature, Tc-To: gelatinization temperature range, ΔH: enthalpy of gelatinization; RC: Relative crystallinity; NCPS: Native cowpea starch; MICPS: Moisture conditioned infrared heated cowpea starch; NCOS: Native corn starch; MICOS: Moisture conditioned infrared heated corn starch, db: dry basis.

Infrared heating of moisture-conditioned cowpea starch also significantly altered the trough viscosity, setback viscosity and pasting temperature (Table 2), but no significant differences were observed in the swelling power, time to peak, peak, breakdown and final viscosities. Cowpea starch infrared heated at a moisture level of 10 g and time of 60 mins showed the highest trough viscosity (423 cP), while the sample infrared heated at 25 g moisture level for 5.68 mins showed the lowest trough viscosity (23 cP). Trough viscosity is the minimum viscosity at constant temperature phase of the pasting and represents the ability of the paste to withstand breakdown during cooling [32]. Except for cowpea starch that was infrared heated

Table 2. Functional and pasting properties of moisture conditioned and infrared heated cowpea starch at different experimental runs*.

Functional and pasting properties of moisture conditioned and infrared heated cowpea starch at different experimental runs*.

Experimental run	Moisture (g)	Time (mins)	WAC (g/g)	OAC (g/g)	SP@ 70 °C	PV (cP)	TV (cP)	BV (cP)	FV (cP)	SV (cP)	Pt (mins)	PT (°C)
1	46.21	37.50	$0.98^{a} \pm 0.02$	$0.52^{c} \pm 0.02$	$3.38^{a} \pm 0.03$	3952^{a}	24^{d}	3929^{a}	4707^{ab}	4684^{b}	7^{a}	74.95^{a}
2	3.79	37.50	$0.88^{cd} \pm 0.01$	$0.63^{bc} \pm 0.05$	$3.37^{a} \pm 0.03$	3825^{a}	29^{d}	3796^{a}	4558^{b}	4529^{b}	7^{a}	74.73^{a}
3	25.00	69.32	$0.95^{abc} \pm 0.06$	$0.57^{abc} \pm 0.01$	$3.39^{a} \pm 0.04$	4048^{a}	30^{d}	4018^{a}	4510^{b}	4480^{b}	7^{a}	74.50^{a}
4	25.00	37.50	$0.88^{cd} \pm 0.04$	$0.53^{c} \pm 0.04$	$3.53^{a} \pm 0.14$	4225^{a}	28^{d}	4198^{a}	4740^{ab}	4713^{b}	7^{a}	74.73^{a}
5	25.00	37.50	$0.91^{abcd} + 0.05$	$0.59^{abc} + 0.01$	$3.33^{a} + 0.42$	3778^{a}	22^{d}	3757^{a}	4534^{b}	4512^{b}	7^{a}	75.25^{a}
6	25.00	5.68	$0.94^{abc} \pm 0.06$	$0.62^{ab} \pm 0.03$	$3.38^{a} \pm 0.42$	3829^{a}	23^{d}	3807^{a}	4591^{b}	4568^{b}	7^{a}	74.73^{a}
7	25.00	37.50	$0.92^{abcd} + 0.07$	$0.66^{a} \pm 0.01$	$3.28^{a} \pm 0.03$	3749^{a}	28^{d}	3721^{a}	4602^{b}	4574^{b}	7^{a}	74.38^{a}
8	25.00	37.50	$0.90^{abcd} \pm 0.03$	$0.65^{ab} \pm 0.03$	$3.23^{a} \pm 0.17$	3743^{a}	27^{d}	3716^{a}	4729^{ab}	4703^{b}	7^{a}	75.15^{a}
9	40.00	15.00	$0.97^{abc} \pm 0.04$	$0.66^{a} \pm 0.06$	$3.35^{a} \pm 0.06$	4344^{a}	129^{c}	4265^{a}	4720^{ab}	4641^{b}	7^{a}	72.68^{a}
10	10.00	15.00	$0.84^{d} \pm 0.03$	$0.58^{abc} \pm 0.03$	$3.29^{a} \pm 0.03$	4165^{a}	358^{b}	3957^{a}	4909^{ab}	4702^{b}	7^{a}	60.25^{b}
11	40.00	60.00	$0.99^{a} \pm 0.01$	$0.65^{ab} \pm 0.06$	$3.41^{a} \pm 0.08$	4402^{a}	127^{c}	4276^{a}	5313^{a}	5187^{a}	7^{a}	62.13^{ab}
12	25.00	37.50	$0.88^{cd} + 0.02$	$0.65^{ab} + 0.05$	$3.43^{a} + 0.08$	3975^{a}	104^{c}	3908^{a}	4942^{ab}	4875^{b}	7^{a}	62.53^{ab}
13	10.00	60.00	$0.90^{abcd} \pm 0.01$	$0.58^{bc} \pm 0.04$	$3.54^{a} \pm 0.04$	4287^{a}	423^{a}	4064^{a}	4839^{ab}	4616^{b}	7^{a}	62.20^{ab}
Native	–	–	$0.89^{bcd} + 0.01$	$0.56^{bc} + 0.05$	$3.34^{a} + 0.48$	3929^{a}	116^{c}	3864^{a}	4800^{ab}	4734^{b}	7^{a}	60.13^{b}

*Values are duplicate determinations for duplicate experimental runs. Mean ± SD. Mean with the same superscript along a column are not significantly different ($p \leq 0.05$)
WAC: Water absorption capacity; OAC: Oil absorption capacity; SP: Swelling power; PV: Peak viscosity; TV: Trough viscosity; BV: Breakdown viscosity; SV: Setback viscosity; Pt: Peak time; PT: Pasting temperature.

at a moisture level of 40 g, all other infrared heated samples generally showed lower setback viscosities than the native cowpea starch (Table 2). Infrared heated cowpea starch samples also showed higher pasting temperatures (60.25–75.25 °C) than the native starch with a value of 60.13 °C (Table 2). The higher pasting temperatures of moisture-conditioned and infrared heated cowpea starches suggests a high resistance to rupture during pasting. A shift to a higher pasting temperature after starch modification has been associated with the damage of starch crystalline region, with a possible increase in the amylose fraction [31]. This is in agreement with the increase in amylose content of the modified starches (Table 3). Through nuclear magnetic resonance (NMR) spectroscopy, Shen [33] found an increase in the amorphous region in potato starch after modification with microwave heating. Furthermore, the increase in pasting temperature also suggest a possible alteration in the chain length distribution of the amylopectin component of the cowpea starch after infrared heat treatment.

Table 3. Composition, relative crystallinity, FTIR peak ratio and thermal properties of native and moisture conditioned and infrared heated starches.

Parameters	NCPS	MICPS	NCOS	MICOS
Moisture (%)	$6.54^{c} \pm 0.28$	$8.72^{b} \pm 0.37$	$10.18^{a} \pm 0.12$	$10.27^{a} \pm 0.28$
Fat (%)	$0.33^{a} \pm 0.01$	$0.25^{b} \pm 0.01$	$0.32^{a} \pm 0.01$	$0.25^{b} \pm 0.01$
Ash (%)	$0.40^{a} \pm 0.03$	$0.42^{a} \pm 0.01$	$0.23^{b} \pm 0.02$	$0.23^{b} \pm 0.01$
Protein (%)	$0.16^{a} \pm 0.01$	$0.17^{a} \pm 0.01$	$0.10^{a} \pm 0.03$	$0.15^{a} \pm 0.04$
Total starch (db %)	$92.58^{a} \pm 0.25$	$90.46^{b} \pm 0.36$	$89.19^{c} \pm 0.06$	$89.11^{c} \pm 0.35$
Amylose (%)	$29.13^{a} \pm 0.22$	$31.67^{a} \pm 2.14$	$20.18^{c} \pm 0.70$	$24.22^{b} \pm 0.25$
RC (%)	$31.91^{c} \pm 0.44$	$31.23^{c} \pm 0.48$	$41.39^{a} \pm 0.25$	$36.18^{b} \pm 1.19$
Ratio of 1045/1022 cm^{-1}	$0.99^{c} \pm 0.01$	$0.97^{d} \pm 0.00$	$1.06^{a} \pm 0.02$	$1.04^{b} \pm 0.01$
T_o (°C)	$69.51^{a} \pm 1.54$	$67.44^{b} \pm 1.54$	$69.42^{a} \pm 0.37$	$69.64^{a} \pm 0.17$
T_p (°C)	$75.79^{a} \pm 0.57$	$72.53^{c} \pm 0.44$	$74.75^{b} \pm 0.51$	$74.89^{b} \pm 0.39$
T_c (°C)	$84.69^{a} \pm 1.13$	$78.59^{b} \pm 2.35$	$80.61^{b} \pm 1.14$	$80.54^{b} \pm 0.28$
T_c-T_o (°C)	$15.18^{a} \pm 2.40$	$11.14^{b} \pm 2.48$	$11.17^{b} \pm 0.82$	$10.90^{b} \pm 0.25$
ΔH (J/g)	$9.38^{b} \pm 1.19$	$11.64^{a} \pm 1.16$	$11.52^{a} \pm 0.89$	$11.88^{a} \pm 1.03$

Mean ± SD. Mean with the same superscript along a row are not significantly different ($p \leq 0.05$).

3.2. Numerical optimization of moisture and infrared heating conditions

Optimization of processing conditions used during the infrared heating of moisture conditioned cowpea starch samples was carried out by maximizing or minimizing responses as explained in Section 2.6 and defining these preset conditions into Minitab 17.0 software (Minitab Inc., State College, PA, USA). Values of responses obtained (Table 2) were input into the software and processed at their set maximum and minimum conditions. Regression coefficients ($\beta_0 - \beta_{12}$) for each of the responses, as well as their R^2 values, as determined by the model used, are presented in Table S1. Optimum conditions of moisture content and infrared heating time obtained were 46.21 g and 32.88 min, respectively with a desirability

factor (D_f) of 0.60. Thereafter, moisture-conditioned, infrared heated cowpea starch (MICPS) and infrared heated corn starch (MICOS) were produced at these optimized moisture and time conditions and then compared with native cowpea starch (NCPS) and native corn starch (NCOS) by determining different parameters such as morphology, crystalline characteristics, functional, thermal and pasting properties.

3.3. Morphology and composition

The scanning electron micrographs (SEM) of cowpea starch and corn starch before and after moisture conditioning, as well as infrared heating treatment are presented in Fig. 1(A–D). Native cowpea starch (NCPS) was predominantly oval-shaped granules with few granules round and irregular in shape. On the other hand, native corn starch (NCOS) appeared irregularly shaped with a few of the granules polygonal while some were round. Furthermore, NCPS had smooth granules with no holes or fissures on the surface compared to the NCOS which were not very smooth. The granules of NCPS are approximately 10–25 μm in diameter, which appeared bigger than the NCOS (6–18 μm). The size and shape of the two native starches in this study are similar to previous reports on cowpea [1], [4], [7], [34] and corn starches [35], [36].

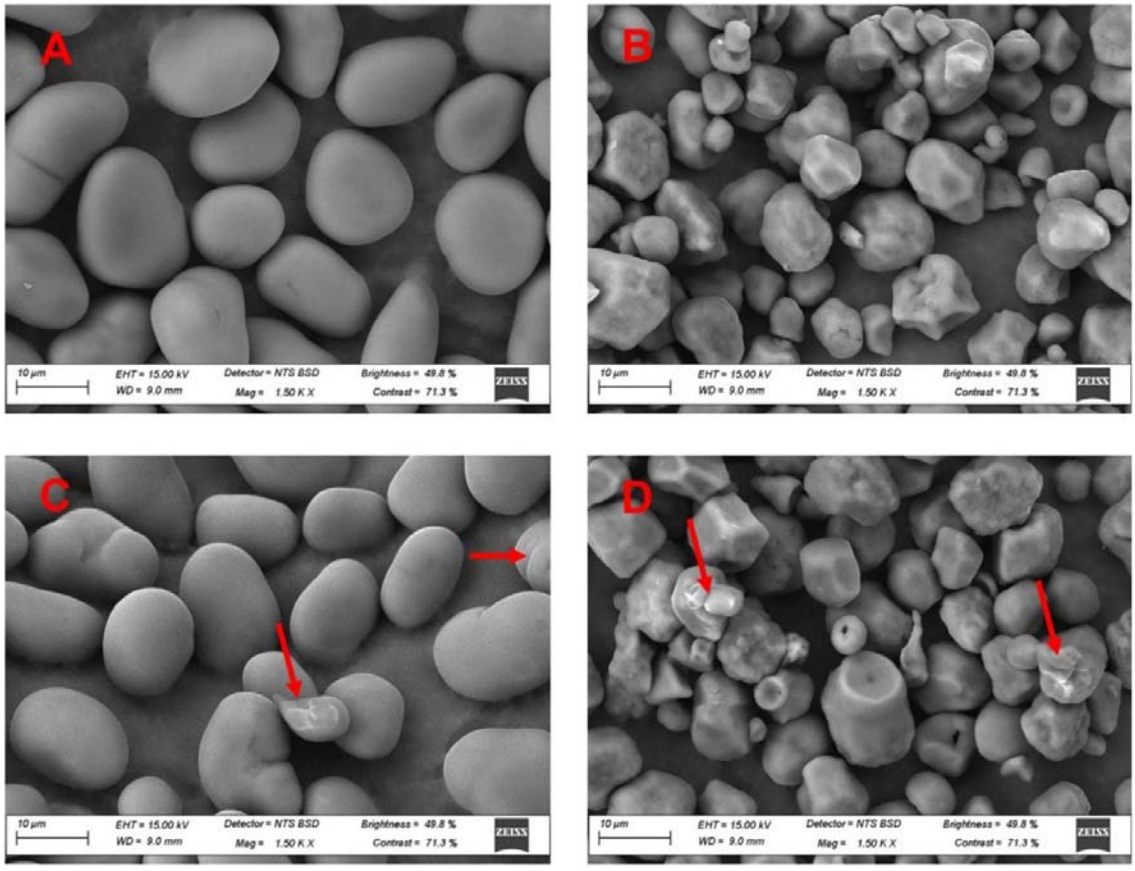


Fig. 1. Scanning electron micrographs of native and moisture conditioned and infrared heated starches

A: Native cowpea starch; B: Native corn starch; C: moisture conditioned and infrared heated cowpea starch; D: moisture conditioned and infrared heated corn starch

Arrows indicate clumping of starch granules.

Infrared heating resulted in slight changes in the morphology of moisture conditioned cowpea starch with slight indentations and clumping observed on the granules (Fig. 1C). However, the changes seen on the corn starch were very substantial. The starch granules showed more clumping together with greater indentations compared with the cowpea starch (Fig. 1D). An earlier study by Mwangwela [6] reported that starch isolated from moisture-preconditioned then infrared heated cowpea at 170 °C did not show birefringence, suggesting that infrared heat treatment resulted in gelatinization of the starch granules. In this study, starch was isolated from cowpea grains before infrared heating at 100 °C and this temperature variation compared with the 170 °C used by early authors may explain the slight changes in granule morphology. Furthermore, the greater surface damage observed in the corn starch granules after infrared heat treatment may be associated with its smaller size compared to the cowpea starch, due to the increased surface area. Changes in granule morphology have also been attributed to leaching of amylose from starch, loosening of the amylopectin crystalline region during heating and re-association of the starch chains within the granules [7], [37]. In this study, the possibility of amylose leaching is ruled out since the starch morphology was done on dried starches. Hence, the loosening of the amylopectin crystalline region during heating and re-association of the starch chains within the granules may explain the changes in granule morphology.

The starch composition including moisture, ash, crude fat, crude protein, amylose content of native cowpea starch, native corn starch and the infrared heated samples are presented in Table 3. All the non-starch components such as ash (0.23–0.42%), crude fat (025–0.33) and crude protein (0.10–0.17%) were generally low indicating that the extracted starches are relatively pure. Furthermore, the total starch (TS) content of cowpea starch (average of 92%) was slightly but significantly higher than that of corn starch (average of 89%). The infrared heating process resulted in a slight reduction in the TS content of the starches and this observation agrees with earlier findings on microwave heat treated lotus seed starch [38].

Native cowpea starch showed significantly higher amylose content (29.13%) than the corn starch reference sample (20.18%). The same trend was observed for the infrared heated cowpea (31.67%) and corn starch samples (24.22%). Except for high amylose corn or maize starches, legume starches generally contain a high level of amylose compared to cereal starches. This property as well as the faster rate of retrogradation and resistance to shear-thinning has increased the demand for legume starch use in different food formulations [39]. From this study, modified starches showed higher amylose content than native starches due to the infrared heat treatment. An increase in the amylose content of the starches after heating suggest partial degradation of amylopectin in cowpea starch. Physical modification methods of starch such as autoclaving-cooling cycles [7] and microwave heating [16], [17], [31] have also been reported to result in total or partial damage of the crystalline region. The amylose contents of native and modified cowpea starches in this study are in agreement with the literature on legume starch [9].

3.4. Crystallinity pattern

The X-ray diffractogram (XRD) patterns of native and infrared-heated starch samples are presented in Fig. 2. Starch may exhibit different crystalline patterns (A, B or C), depending on the botanical origin and the arrangement of the crystalline lamellae of amylopectin. The NCPS and NCOS both exhibited strong peaks at 15° (2θ), a connected doublet at 17° and 18° (2θ), a single peak at 23° (2θ) and minor peaks at approximately 11, 12 and 27° (2θ). Additionally, the NCOS showed another weak peak at 20° (2θ), suggesting that the NCOS

corresponds to the A-type crystallinity. Cereal starches including corn starch are known to display the A-type crystallinity, while legumes starches generally exhibit the C-type pattern. However, C-type starches can be further classified into C_A- type, which is closer to A-type and C_B-type which is closer to B-type polymorphs [4], [40]. Thus, the NCPS can be classified as C_A-type crystallinity since it is closer to the A-type polymorph than the B-type (Fig. 2).

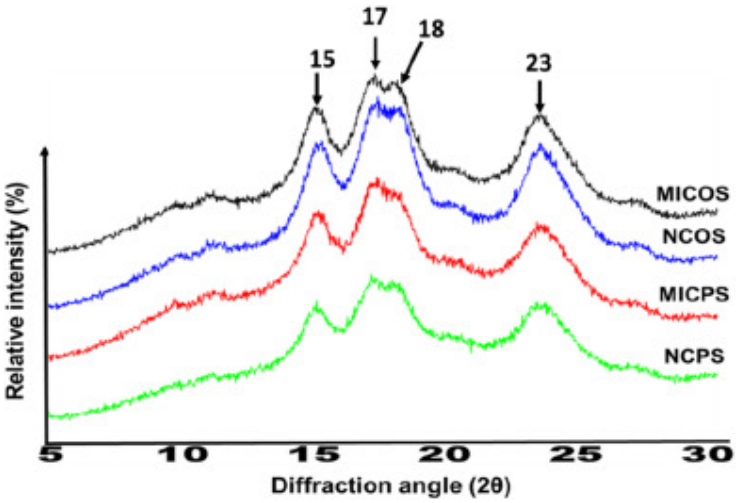


Fig. 2. X-ray diffraction patterns of native and moisture conditioned and infrared heated starches

NCPS: Native cowpea starch; MICPS: moisture conditioned and infrared heated cowpea starch

NCOS: Native corn starch; MICOS: moisture conditioned and infrared heated corn starch.

Infrared heat treatment did not change the crystallinity patterns of moisture conditioned cowpea and corn starches (Fig. 2). However, infrared heating resulted in a decrease in the relative crystallinity (RC) of the starch samples, although the decrease was not significant ($p > 0.05$) for cowpea starch (Table 3). The RC result for cowpea and corn starches are in agreement with their amylose content, which was also not significant ($p > 0.05$) for cowpea starch but significant for corn starch after modification (Table 3). The variation in packing arrangement of the starches as well as their level of hydration may have influenced their behaviour during infrared heating. It has been reported that the A-type starch crystallites have double helices that are closely packed with only four inter-helical water molecules and the centre of lattice contains a pair of double helices, while the B-type are more hydrated with 36 inter-helical water molecules [41], [42]. Another plausible reason for the variation in the RC could be associated with differences in the amylose contents of the starches (Table 3). The amylopectin molecules form the crystalline structure in starch granules, therefore, it is expected that RC will be inversely related to amylose content [43], [44]. Furthermore, the infrared heating process may have affected the crystalline region resulting in a partial loss of the crystalline array due to the breakdown of hydrogen bonds in the starch granules. A similar observation has been reported for starches subjected to microwave heating [16], [45]. Yang [16] using Nuclear magnetic resonance showed that the α-(1,6) glycosidic bonds were destroyed more easily than α-(1,4) glycosidic linkages during microwave treatment of waxy maize starch. Since the α-(1,6) glycosidic bonds are found in the amylopectin component of starch and this starch molecule forms the crystalline region of starch, then it is likely that the infrared heating may also have resulted in greater disruption of this region in the corn starch granules as shown by the reduction in RC (Table 3). Short A amylopectin chains (DP 6–12)

have been suggested to contribute more to starch crystallinity [46] and a decrease in the proportion of this short-chain has been attributed to a damage of the crystalline region with a resultant decrease in RC [16], [47]. Hence, the insignificant decrease in RC for cowpea starch suggest the short chains (A chain) in cowpea starch are more resilient than those in corn starch. Future studies are required to validate this claim.

3.5. FTIR

The FTIR spectra for native and modified starches were similar and indicates the polysaccharide nature of the starch samples (Fig. 3). Cowpea starch (native and infrared-heated) showed sharper peaks in the broadband region of 3000–3650 cm^{-1} compared to corn starch, which was broader. This broadband region has been attributed to the complex vibration stretching of free, inter-and intra- molecular hydroxyl (-OH) groups [48], [49]. The corn starch control showed higher intensities than the cowpea starch in the OH-region suggesting a higher density of strong hydrogen bonding interactions in corn starch [49], [50].

The starches also showed an absorption peak at around 2931 cm^{-1} contributed by the C H stretching associated with the ring methine hydrogen atoms [48], while a similar peak found in the region of 1650 cm^{-1} could be due to the bending vibration of bound water in the amorphous region of starch [51], [52]. Furthermore, the starch samples exhibited a sharp peak at around 995 cm^{-1} in the fingerprint region which denotes the vibrations of the glucose C-O-C bond in the starch structure [53], [54].

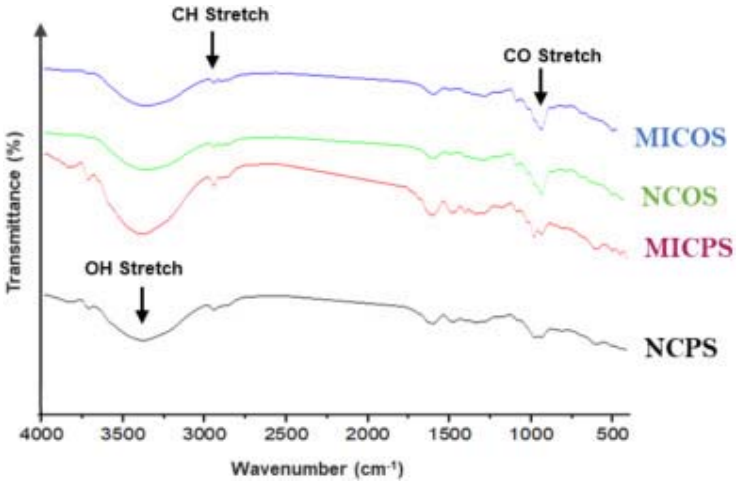


Fig. 3. FTIR spectra of native and moisture conditioned and infrared heated starches

OAC: Oil absorption capacity; WAC: Water absorption capacity

NCPS: Native cowpea starch; MICPS: Moisture conditioned infrared heated cowpea starch

NCOS: Native corn starch; MICOS: Moisture conditioned infrared heated corn starch.

To assess the impact of infrared heating on the short-range structure of the starches, the bands at 1045 and 1022 cm^{-1} which are linked with the crystalline and amorphous regions in starch, respectively were determined and their ratio reported in Table 3. The ratio of these bands (1045/1022 cm^{-1}) is frequently used to quantify the degree of crystalline order in starch samples [55], [56]. Infrared heating increased the peak intensity at 1045 and 1022 cm^{-1} for

cowpea starch but decreased the same peaks for corn starch. Furthermore, infrared-heated starches exhibited lower 1045/1022 cm^{-1} ratios than the native starch samples (Table 3) indicating that infrared heating resulted in the reduction of the double-helical order structure of the starch granules in the external region [57]. The reduction in the 1045/1022 cm^{-1} ratios which suggest a reduction in the degree of crystallinity is consistent with the XRD results (Fig. 2) and RC data (Table 3).

3.6. Water and oil absorption capacities

Native corn starch showed significantly ($p \leq 0.05$) higher WAC than the cowpea starch (Fig. 4). The same trend was observed for the moisture conditioned infrared heated starch samples. The relatively higher WAC of native and modified corn starch compared with the native and modified cowpea starch may be associated with the differences in their crystalline order as shown by the FTIR data which was higher in corn than in cowpea (Table 3). This observation is also consistent with the RC data (Table 3). The infrared heating process though did not significantly ($p > 0.05$) change the ability of the starch samples to absorb oil, the WAC of the cowpea starch and the reference corn starch sample slightly increased after infrared heat treatment (Fig. 4). While the difference in starch granule size (Fig. 1) may explain the variation in WAC for native starch samples, the slight variation observed in the WAC after infrared heating suggest varying levels of re-arrangement within the amorphous region of the respective starch samples. It has been suggested that differences in intragranular molecular rearrangement can lead to varied accessibility of water to the amorphous regions of starch [58], [59]. The increase in WAC of the starches after modification may be associated with the increase in amylose content of the starches (Table 3) since this starch component (amylose) is known to greatly influence WAC [23]. Earlier researchers also found that legume starches with relatively high amylose content displayed high WAC [23], [30]. As previously noted (Section 3.1), the increase in WAC may also be partly due to the formation of simpler molecules such as sugars and dextrins due to starch depolymerization. These smaller molecules have more affinity for water absorption than starch and such changes have been reported for microwaved heated [31] and gamma-irradiated legume starches [13].

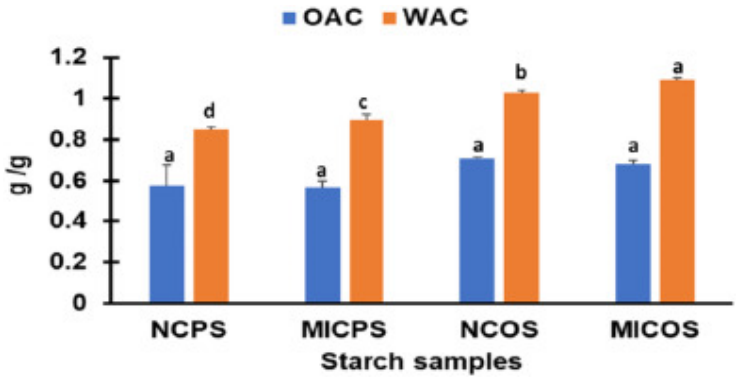


Fig. 4. Oil and water absorption capacities of native and moisture conditioned and infrared heated starches

OAC: Oil absorption capacity; WAC: Water absorption capacity

NCPS: Native cowpea starch; MICPS: Moisture conditioned infrared heated cowpea starch NCOS: Native corn starch; MICOS: Moisture conditioned infrared heated corn starch

Error bars indicate standard deviation (N = 4)

Bars with same different alphabest indicate significant difeerences ($p \leq 0.05$).

3.7. Swelling power

The swelling power of cowpea and corn starches generally increased with an increase in temperature (Fig. 5). Native cowpea starch showed substantially higher swelling power than the native corn starch. Swelling behaviour results from the ability of starch to trap and retain water within its structures before and during gelatinization. The variation in swelling power of starches has been linked with differences in the ratio of amylose to amylopectin. According to Tester and Morrison [60], the amylopectin fraction of starch is primarily responsible for swelling, while amylose has been suggested to restrict starch swelling behaviour [27]. Hence, starches with low amylose content would display high swelling power. In this study, the inverse relationship of amylose with swelling power was only true at lower temperature of 50 and 60 °C (Fig. 5). At higher temperatures (70-90 °C), cowpea starch with a relatively higher amylose content displayed greater swelling, indicating other factors are controlling the swelling behaviour of these starches at higher temperatures. Some other researchers have also reported that starches with significantly high amylose content did not show restricted swelling [44], [61]. Besides the ratio of amylose to amylopectin, the presence of endogenous lipids in corn starch has also been reported to restrict starch granular swelling [60]. Furthermore, variation in the chain length distribution of amylopectin component of the starches may also explain the differences in the swelling power. For example, Pearson correlation analysis of starch from nine genotypes of quinoa showed that more short internal chains of amylopectin are highly correlated to a higher swelling power [62]. According to their report, the presence of short internal chains of amylopectin contributed to a more disordered packing of double helices in the starch granules, resulting in easier swelling and water hydration.

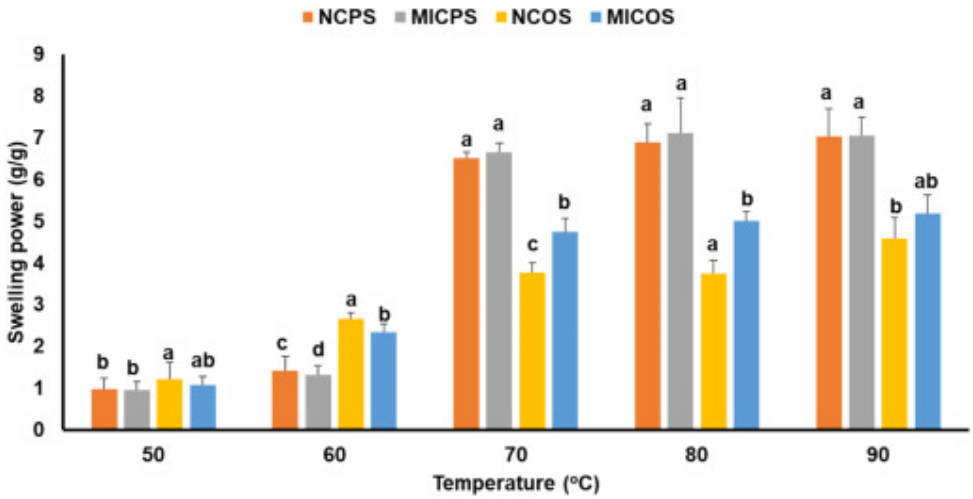


Fig. 5. Swelling power of native and moisture conditioned and infrared heated starches

NCPS: Native cowpea starch; MICPS: Moisture conditioned infrared heated cowpea starch

NCOS: Native corn starch; MICOS: Moisture conditioned infrared heated corn starch

Error bars indicate standard deviation (N = 4). Bars with same different alphabest indicate significant difeerences ($p \leq 0.05$).

Infrared heating generally increased the swelling power of both starches, but the increase was marginal in cowpea starch (Fig. 5). The varied impact of infrared heating time on the swelling ability of the starches could be attributed to the nature of the starch granules, with regards to size and compactness (intragranular arrangement). Cowpea starch appeared bigger (Fig. 1) and possibly have a less compact granule with higher intermolecular areas than the corn starch. This presumably reduced the effect of infrared heating on the swelling ability of the cowpea starch. Previous studies indicated that the greater the swelling capacity of starch granules, the weaker is the binding forces within the granules [63].

3.8. Thermal properties

Native cowpea and corn showed similar onset gelatinization temperature (T_o) of approximately 70 °C (Table 3). However, the peak gelatinization temperature (T_p), conclusion gelatinization temperature (T_c), and gelatinization temperature range (T_c-T_o) of the native cowpea starch were significantly ($p \leq 0.05$) different from that of the native corn starch. Infrared heating significantly decreased the T_o, T_p, T_c, and T_c-T_o of the cowpea starch sample. Meanwhile, the modification process did not significantly alter these parameters in the corn starch control. The decrease in the gelatinization temperatures and gelatinization range of the cowpea starch indicates that moisture conditioned, and infrared heated starch sample gelatinized faster by losing their integrity, which presumably results from the weakening of the starch granule structure (Fig. 3). This may explain why the swelling power (Fig. 5) and peak viscosity (Table 3) of the starches increased after infrared heat treatment. Starch gelatinization temperatures have been reported to decrease with an increase in starch damage [63]. Previous studies similarly found a reduction in the T_p of flour from moisture-preconditioned infrared heated Bambara groundnut [19] and cowpea [6]. The gelatinization temperature for cowpea starch in this study is in agreement with values (69.60 and 89.00 °C) reported in the literature [4], [6], [7], [39], [64], [65]. The influence of amylose content and amylopectin structure on the thermal properties have been very contradictory. While some authors reported a positive correlation of amylose content and amylopectin long chain with gelatinization temperatures [66], other authors found a significant negative correlation of amylopectin short with gelatinization temperatures [67], [68]. However, it has been established that starch gelatinization temperatures depend largely on the distribution of amylopectin short chains rather than the ratio of amylose to amylopectin [69]. Thus, future studies may be required to establish the role of amylopectin chain length on the thermal properties of infrared heated starches.

The enthalpy of gelatinization (ΔH) of native cowpea starch (9.38 J/g) was lower than that of native corn starch (11.64 J/g) (Table 3). The ΔH value is a measure of the overall crystallinity of starch and is an indicator of the loss of molecular order within the starch granules during gelatinization [63]. It is influenced by the degree of crystallinity, intermolecular bonding, genetic and environmental factors [70].

3.9. Pasting properties

The pasting properties of native and modified starches are shown in Table 4. Corn starch showed significantly ($p \leq 0.05$) higher peak, breakdown, final and setback viscosities than cowpea starch. The higher peak viscosity displayed by the corn starch could be due to it higher degree of crystallinity (Table 3) and lower amylose content compared to cowpea starch (Table 3). Starches with high amylose contents would show low peak viscosity due to restricted swelling of starch granules during pasting [71]. The variation in starch granule size

(Fig. 1) may also influence the peak viscosity of the starches (Table 4). Small-sized starch granules will absorb more water and swell due to increased surface area and starch volume. The extent of damaged starch content between the corn and cowpea starches could also explain the variation in their peak viscosities. Wang and Wang [72] noted that damaged starch content plays a more important role in determining the peak viscosity of starch, while damaged starch was negatively correlated with peak viscosity of rice starch [73]. The crystalline domains of the starch granules are due to the clustered branches of amylopectin chains that are packed together. The peak viscosity, which is a measure of the swelling power of the starch in terms of the resistance of swollen granules to shear may be influenced by the ratio of amylose to amylopectin [71], [74] and the chain length distribution of amylopectin [74]. A correlation analysis showed that the pasting properties of starch were affected by the fine structures of both amylose and amylopectin [75]. Infrared heating did not significantly alter the pasting properties of corn starch, although the peak, final and setback viscosities increased. However, the cowpea starch samples showed a significant ($p \leq 0.05$) increase in the peak, breakdown and final viscosities (Table 4). The increase in peak viscosity after infrared heating presumably results from the loosening of the starch structure allowing for greater mobility of water and swelling during pasting. It is hypothesized that the infrared heat treatment may have resulted in some level of depolymerisation of the starch granules at the molecular level, which may explain the reduction in the 1045/1022 cm^{-1} ratios (Table 3). The peaks in these regions are known to be sensitive to changes in the structure of the starch at the granule surface. The result of the peak viscosity is in agreement with the WAC (Fig. 4) and swelling power (Fig. 5), which also increased after infrared heating. Furthermore, the contribution of the chain length of amylose [76], [77] and distribution of amylopectin chains [74], [78] cannot be ruled out.

Table 4. Pasting properties of native and moisture conditioned and infrared heated starches.

Parameters	NCPS	MICPS	NCOS	MICOS
Peak viscosity (cP)	3929.00^{c}	5829.00^{b}	7617.50^{a}	7808.50^{a}
Breakdown viscosity (cP)	3864.00^{c}	5745.00^{b}	6559.50^{a}	6293.00^{a}
Final viscosity (cP)	4800.00^{d}	5331.50^{c}	6629.00^{b}	7148.50^{a}
Setback viscosity (cP)	4734.00^{c}	5247.50^{b}	5586.00^{a}	5633.00^{a}
Pasting temperature (°C)	60.13^{c}	72.50^{a}	70.23^{b}	70.22^{b}

Values are mean of duplicate determinations. Mean with the same superscript along a row are not significantly different ($p \leq 0.05$).

NCPS: Native cowpea starch; MICPS: Moisture conditioned infrared heated cowpea starch.

NCOS: Native corn starch; MICOS: Moisture conditioned infrared heated corn starch.

Previous studies though reported an increase in pasting viscosity of pre-conditioned cowpea and Bambara groundnut flour after infrared heating [6], [19]. However, it should be noted that the current study extracted starch from cowpea before infrared heat treatment. Hence, the difference in peak viscosity results may be attributed to the presence of other non-starch components such as fibre, protein and fats in the flour, which may restrict starch granule swelling during pasting.

The pasting temperature of cowpea starch significantly increased after infrared heating but that of corn starch showed a slight insignificant decrease (Table 4). Pasting temperature represents the temperature at which the starch cooks and is very important in determining the

temperature requirements in industrial applications. Higher pasting temperature indicates greater resistance of starch granule to swelling [39]. The pasting temperature of starch is influenced by the amylose content, structure of amylopectin and the prevailing conditions during pasting measurement [79].

High amylose starches have been shown to display higher pasting temperature and lower peak viscosity while those with lower amylose content have higher swelling power and less tendency to retrograde [74]. Furthermore, starches with higher amylose content and a greater proportion of amylopectin long chains have reportedly displayed higher starch pasting temperature, decreased peak viscosity and shear thinning [66], [74] and this was the case in this study, except that the chain length distribution of the amylopectin was not determined. The pasting temperature values observed for native and modified cowpea starch in this study is in agreement with the literature [1], [4], [39], [78].

4. Conclusion

Optimization of moisture level and infrared heating time for cowpea starch modification showed that the optimum moisture level and micronizing time were 46.21 g and 32.88 mins, respectively. Infrared heating under optimized conditions improved the swelling power, water absorption, and pasting properties of cowpea starch. Modified cowpea starch, however, exhibited a higher swelling power, but lower water absorption capacities and lower pasting properties compared with corn starch control. The modified cowpea starch had reduced crystallinity and a change in the double-helical order structure as evident in the XRD and FTIR data, respectively. The infrared heat treatment is a novel and promising physical modification method for improving the physicochemical properties of cowpea starch. Future studies are required to determine the impact of the infrared heating on the chain length distribution of amylopectin fraction of the starch and the level of depolymerization using NMR techniques as well as the application of the starch in foods systems that require high swelling properties.

CRediT authorship contribution statement

Samson A. Oyeyinka: Research conceptualization, data curation, analysis and draft manuscript writing.

Ajibola B. Oyedeji: Research conceptualization, data curation, analysis, draft manuscript writing.

Opeolu M. Ogundele: Research conceptualization, data curation, analysis, reading of draft manuscript writing.

Oluwafemi A. Adebo: Reading of draft manuscript, validation and funding.

Eugénie Kayitesi: Conceptualization, reading of draft manuscript, validation and funding.

Patrick B. Njobeh: Supervision, reading of draft manuscript, validation and funding.

Declaration of competing interest

We have no conflict of interest to declare.

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