Effect of chemical treatments on the functional, morphological and rheological properties of starch isolated from pigeon pea (*Cajanus cajan*)

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ABSTRACT

Different chemical treatments (cross-linking, oxidation, and hydroxypropylation) were used to modify pigeon pea starch, and its effect on physiochemical, pasting and rheological properties were studied. Cross-linking and oxidation decreased while hydroxypropylation increased the swelling power of pigeon pea starch. All starch samples showed a decrease in their paste clarities. FTIR spectra of all starch samples displayed characteristic absorption bands of starch at wave numbers 1076, 1148, 1376, and 1632 cm^{-1}. A significant reduction occurred in peak, cold paste, hot paste, and setback viscosity after chemical modification. Rheological determinations showed that starch pastes had viscoelastic behaviour. G′ and G″ of all starch paste increased after chemical modification. Native and chemically treated starches revealed oval to elliptical-shaped granules and no change was observed after modification when examined in SEM. These results confirmed that the undesirable properties of native pigeon starch can be suitably altered via chemical treatments to make them suitable for several food applications.

1. Introduction

Pigeon pea (*Cajanus cajan*) is a perennial legume also known as red gram or locally called *Arhar Dhal* and grown all over Africa and Asia for human food and animal feed. Pigeon pea seeds contain about 39–58.9% starch, 11.62–18.17% resistant starch, and 20–24% proteins (Emefiene et al., 2014). The separation of starch from various pulses is very difficult due to the presence of insoluble protein and fiber (Esan and Ojemola, 2018). Native starches have a limited number of industrial applications due to some undesirable characteristics such as high retrogradation rate, low shear stress resistance, high viscosity, low light transmittance, low decomposition rate, and low solubility, (Yadav et al., 2013). Native legume starches have a generally high retrogradation rate and tend to undergo syneresis. Therefore, the requirement of modification in native starches to enhance their ideal functional properties is required for their industrial applications. The chemical modification offers an easy approach to overcome the drawbacks of native starches as they bring many changes in the starch physicochemical properties through the insertion of the different functional groups onto the starch molecules (Goswami et al., 2018) using reactions of derivatization (etherification, esterification, and cross-linking) or decomposition (oxidation and acid hydrolysis) (Naknaen et al., 2017). Cross-linked

starches are generally made with a cross-linking reagent either by the use of STPP/STMP or epichlorohydrin. Oxidized starches are commonly prepared with sodium hypochlorite (Naknaen et al., 2017) and hydroxypropylated starches are made with propylene oxide added to the native starch in the presence of an alkaline catalyst (Hjermstad, 1971). Starches modified with oxidation, cross-linking, and hydroxypropylation have improved durability, storage stability at low temperatures, freeze-thaw stability, paste clarity, reduce gelatinization temperature, film formation, and adhesion properties (Lawal, 2011).

Chemical modification techniques such as cross-linking, oxidation, and substitution are used to produce thickeners with the required rheological properties during storage and shipment (Moore et al., 1984). Rheological studies help to understand the molecular structure or distribution of foods' molecular components, as well as the estimation and characterization of structural changes in foods during their manufacturing processes (Koocheki and Razavi, 2009). As a result, food rheology can offer useful information for product development, quality control, sensory evaluation, and process design. The dynamic rheological assessments for small deformation oscillatory measurements provide valuable information on the viscoelastic properties of starch pastes without causing structural elements to be disturbed (Gunasekaran and Mehmet, 2000). Yousefi and Razavi (2015) investigated the dynamic

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rheological properties of cross-linked and hydroxypropylated wheat starch gels at 8% and 12% w/w concentrations and discovered that the native wheat starch and cross-linked wheat starch samples exhibited an intermediate behavior between a weak and an elastic gel (tan 0.14–0.64), while the hydroxypropylated wheat starch samples exhibited an elastic gel behavior (tan 0.10–0.11), which was unaffected by concentration. They also concluded that hydroxypropylated wheat starch (in terms of viscoelastic properties) and cross-lined wheat starch (in terms of paste properties) have unique dynamic rheological properties that can be used in a variety of gel-like foods.

Many traditional sources of starch are currently available and are being overutilized for a wide range of commercial purposes. Starches from non-conventional sources such as pigeon pea could be used for industrial applications as it is still underutilized legume. However, pigeon pea starch has certain limitations in its native form such as dehulling problems and long cooking time (Esan and Ojemola, 2018) which makes it less important for industrial use. This study also becomes imperative because pigeon pea starch is a cheaper source than most other conventional sources of starch and can be used in industrial and domestic applications apart from food. Such applications include textile, pharmaceuticals, paper, and the development of several other functional materials for polymer technologies (Lawal et al., 2009). The rich carbohydrate content of pigeon peas makes them a reasonable source of starch. Less research work has been carried out on the rheological properties of oxidized, cross-linked, and hydroxypropylated pigeon pea starch. In consideration of this, the purpose of the present study is to evaluate the functional, morphological, and rheological properties of chemically treated pigeon pea starch.

2. Materials and methods

2.1. Materials and reagents

Pigeon pea (*Cajanus cajan*) legume seeds were purchased from the main market at Rohtak, Haryana (India). All types of reagents and chemicals used for research purposes were of analytical grade and purchased from Sigma Chemical Co. (St. Louis, MO, USA).

2.2. Starch isolation

The starch was isolated from pigeon pea seeds using the method of Narina et al. (2012) with slight modifications. The seeds were soaked for 12 h in warm distilled water at room temperature. The seed coat was removed from the softened seeds and then ground to make a fine paste with the addition of distilled water by using a laboratory blender. The 200-mesh sieve was used for screening ground slurry. After this, the filtrate was allowed for 8 h for the complete settling of starch particles gravimetrically. Pure starch was oven-dried at 40 °C overnight and stored in an airtight container for further study.

2.3. Chemical composition of starch

Different constituents such as crude protein, ash, fat content, and moisture content present in native pigeon pea starch were analyzed by using standard methods of AOAC (1995). The amylose content of a starch sample was measured by the method adopted by Hoover and Ratnayake (2001). Before the estimation of amylose content, all starch samples were defatted with hot n-propanol-water (3:1 v/v) for 7 h and dry in a hot air oven for 24 h at 30 °C. 20 mg defatted starch was taken in a capped centrifuge tube containing 8 ml 90% dimethylsulfoxide (DMSO) and mixed for 20 min in a shaker. Then the vial was heated in a water bath for 15 min at a temperature of 85 °C with proper shaking. The sample tube was cool down and diluted with distilled water to make a volume of 5 ml in a volumetric flask. 1 ml of diluted sample was taken with a pipette and mixed with 40 ml of distilled water. Add 5 ml of I_2/KI solution (0.0025M KI) to it and make a final volume of 50 ml with distilled water. The solution was allowed to stand for 15 min and then measured at 600 nm by a double beam spectrophotometer. A standard curve of pure potato amylose and amylopectin was prepared to correct the overestimation of amylose content.

2.4. Cross-linking

Cross-linking of starch was done by the method stated by Chung et al. (2008) with slight modifications. Cross-linked starch was prepared by adding the mixture of STMP and STPP (99/1% w/w, 2g) to the starch slurry. 50 g starch sample was taken in a beaker and 1.98 g STMP and 0.02 g STPP were added with 100 ml of distilled water in it and the pH of the slurry was adjusted to 11 (with 0.1M NaOH) and taken into a hot water bath with continuous stirring for 3 h at 45 °C. The slurry pH was maintained to 11 with 0.1M sodium hydroxide, neutralized with 1M hydrochloric acid, and washed 4–5 times with demineralized water. The obtained wet starch was dried at 40 °C and stored. The degree of cross-linking was determined using the viscosity values obtained by the RVA method suggested by Chung et al. (2008). The degree of cross-linking of cross-linked starch was determined from the viscosity value obtained from the Rapid visco analyzer (RVA). The peak viscosity of modified starch slurry was measured from the RVA results. A programmed heating and cooling cycle to obtain the peak viscosity was employed as follows: Starch slurries were heated from 50 to 95 °C at 11 °C/min, and then held at 95 °C for 2 min afterward, the paste was cooled down to 50 °C at 11 °C/min and finally kept at 50 °C for 2 min.

The degree of cross-linking was calculated by using the equation below:

Degree of cross-linking (%) = (A-B)/A × 100

Where A is the peak viscosity of the control sample (without STMP/STPP), and B is the peak viscosity of the cross-linked starch.

2.5. Oxidation

Oxidized starch was prepared using the method of Forssell et al. (1995) by adding 20 g of NaOCl (4.0 g of active chlorine/100 g of starch resulting in 1.6 g of active chlorine, w/w) drop-wise to the 50% starch slurry (50 g starch in 100 ml distilled water) over 30 min, while maintaining a pH range of 9.0–9.5, with constant stirring at 30 ± 2 °C. The pH of the slurry was adjusted to 9.5 with 2M NaOH. After the addition of NaOCl, the reaction was allowed to continue for 50 min. Now, the pH was adjusted to 7 with 1M HCL, and the starch was filtered 3–4 times with distilled water. Finally, the starch was oven-dried at 45 °C for 12 h to obtain the hypochlorite-oxidized pigeon pea starch. Carboxyl and carbonyl content of oxidized starch was measured by using the method of Chattopadhyay et al. (1997) and Smith (1967).

2.6. Hydroxypropylation

Starch hydroxypropylation was done using the method of Hjermstad (1971) and the degree of substitution present in the hydroxypropylated starch was determined by the standard equation.

Degree of substitution = 162 HP %/(5800–58 HP %)

2.7. Color evaluation

Hunter color Flex EZ 45/0 (Hunter Associates Laboratory, Reston, Virginia, 20190 USA) was used to measure the change in color of native and chemically treated pigeon pea starches. White tile is used for the calibration of the chroma meter and the L* (whiteness) value was obtained.

2.8. Starch solubility and swelling power

Native and chemically treated pigeon pea starch sample's solubility and swelling power were determined using the method of (Lauzon et al., 1995). The starch dispersion (0.5 g starch in 25 ml distilled water) was heated at different temperatures of 55, 65, 75, 85, and 95 °C for 1 h with continuous shaking followed by rapid cooling to room temperature and centrifugation at 1900×g for 15 min. The % solubility and swelling power were calculated as given below

$$\text{Weight of dried sample} = \frac{\text{Weight of dried sample}}{\text{Sample weight}} \times 100$$

$$\% \text{ Swelling power} = \frac{\text{Weight of wet sample}}{\text{Sample weight} \times (100 - \% \text{ solubility})} \times 100$$

2.9. Morphological properties

A scanning electron microscope was used to study the morphological properties of native and chemically treated starch samples. The starch samples were sprinkled over a double-sided silver tape attached to the stub, coated with a thin film of gold palladium, and examined under a scanning electron microscope (Zeiss EVO 50) with 20 kV of accelerating voltage and $1.00\times$ magnification.

2.10. Fourier transform-infrared spectroscopy (FT-IR)

FT-IR spectra of native and chemically treated starches were obtained using the FT-IR spectrometer (Bruker Alpha, Platinium ATR, Germany). The starch samples were directly placed on a sampling unit and the spectra were recorded in the range of $600–4000 \text{ cm}^{-1}$. Absorbance height ratios at $1047/1022 \text{ cm}^{-1}$ were obtained using absorption peak values.

2.11. Paste clarity

Paste clarity of starch was determined by the method adopted by Khan et al. (2014). The starch paste was stored for 5 days at a cooling temperature of 4 °C and % transmittance was noted every 24 h by using a UV–visual double beam spectrophotometer at 650 nm.

2.12. Rapid visco analysis (RVA)

Rapid visco analyser (RVA Techmaster, Perten instruments, Newport Scientific, Australia) was used to measure the RVA characteristics of all starch samples. The starch sample was directly taken into a canister, and 25 ml of distilled water was added to it making a final weight of 28 g (10% w/w). A sample heating and cooling cycle of 13 min was used and the sample was heated from 50 °C to 95 °C over 3.5 min, then held at 95 °C for 2.5 min, cooled from 95 °C to 50 °C over 4 min, and then held at 50 °C for 1 min. Parameters, including pasting temperature, Peak viscosity, breakdown, hot paste viscosity, setback, cold paste viscosity, stability ratio (HPV/PV), and set back ratio (CPV/HPV) were recorded.

2.13. Starch gel hardness

Gel hardness of native and chemically treated starch gels was determined with 2 texture analyzer TA-XTi (Stable Micro Systems, Godalming, UK). Pastes obtained after pasting measurement in a canister were sealed with parafilm to avoid moisture loss and stored in the refrigerator for 24 h at 4 °C. The gel samples were left for 4 h at room temperature before the measurement. The gels were punctured using a cylindrical probe up to a distance of 10.0 mm with a speed of 1.0 mm/s. The gel hardness was determined by measuring the peak force of the first penetration and termed gel hardness.

2.14. Rheological properties

2.14.1. Dynamic properties

The rheological properties of native and chemically treated starch samples were measured with Modular Compact Rheometer (Model-52, Anton Paar, Austria) attached with a parallel plate geometry having a 4 cm diameter in size. During measurements, a 1000 µm gap size was set in the rheometer. For frequency sweep calculations, the starch paste (10 g/100 g) was made and heated at 85 °C in a water bath for 3 min with constant stirring. The sample was cooled and filled on the plate. The frequency range from 0.1 to 100 rad/s at 25 °C was used to perform frequency sweep tests and loss tangent (tan δ), storage modulus (G'), and loss modulus (G") was obtained.

2.14.2. Steady shear measurement

To determine steady shear properties, the starch sample (10 g/100g) was continuously sheared from 1 to 1000 s^{-1}. To identify the change that occurred in the rheological properties of native and chemically treated starch samples under steady shear, the data were fitted to the Herschel-Bulkley model:

$$\sigma = \sigma_o + K(\gamma)^n$$

where σ {shear stress (Pa)}, σ_o (yield stress), γ {shear rate (s^{-1})}, K {consistency index (Pa.s^n)}, *n* (behavior index).

2.15. Statistical analysis

Triplicate results were taken for all samples and the mean values ± SD were obtained. The results were analyzed by using the one-way analysis of variance (ANOVA) method, at a significant difference level ($p < 0.05$). Statistical software (SPSS 19.0) was used for analyses.

3. Results and discussion

3.1. Proximate composition

The native pigeon pea starch had a moisture content of 10.98%, protein content of 0.90%, ash content of 0.03%, and lipid content of 0.79%. The purity of the starch was reflected by the lower percentages of ash, protein, and fat in the starch. The starch yield was found to be 56.56% (w/w, based on the weight of dried seeds). The amylose content of native, oxidized, cross-linked, and hydroxypropylated pigeon pea starch was differed significantly ($p < 0.05$), and values found to be 29.15%, 32.28%, 17.57%, and 33.02%, respectively. The amylose content is the main determinant for starch properties and applications. The amylose content was reduced significantly ($p < 0.05$) after cross-linking and increased after the oxidation and hydroxypropylation treatments. Previously, Fonseca et al. (2015) and Wojeicchowski et al. (2018) reported similar results for an increase in amylose content in oxidized potato starch and common bean starch. The degradation of amylose molecules during the oxidation process causes an increase in amylose content in oxidized starch. In addition to creating carboxyl groups, oxidation would cleave starch molecules. The cleavage of high-molecular-weight amylose molecules would produce some amylose that could be measured (Kuakpetoon and Wang, 2008). A decrease in amylose content in litchi starch cross-linked by STMP was noticed by Sharma et al. (2021) and in white sorghum, starch cross-linked by STMP/STPP by Goswami et al. (2018). The decrease in amylose content in cross-linked starch may be due to intermolecular bonding between amylose and amylopectin molecules or within amylose molecules (Wang and Wang, 2003). The increased substitution in the functional group converts the starch into a highly cross-linked form, which may also account for the decrease in amylose content in cross-linked starch. In hydroxypropylated treatment, the amylose content was significantly ($p < 0.05$), increased and it was due to the

presence of hydroxypropyl groups formed during hydroxypropylation in the starch chains and which are primarily introduced in the amorphous regions composed mainly of amylose (Hood and Mercier, 1978). All these variations in the amylose content of modified starches could be due to the degradation of starch granules arising from modification (Adebowale et al., 2002; Lawal, 2004). The differentiation in amylose content affects the functional and physicochemical properties; starches with higher amylose content produce a stiff and non-transparent gel and make them an important ingredient for making meat sausages and meat emulsions which is highly required in texture improvement. Starches with a high amount of amylose content can be used as a raw material for making edible packaging (Pelissari et al., 2012).

3.2. Degree of modification of starch

The degree of oxidation in oxidized pigeon pea starch granules was determined by the concentration of carbonyl and carboxyl content. After modification, the hypochlorite-oxidized starch contained 1.6 g of active chlorine (w/w) and carbonyl content was observed to be 0.031%. Similar findings for oxidized bean starch are reported by Vanier et al. (2012). The carboxyl content of oxidized pigeon pea starch was about 0.037%. Garcia et al. (2014) also report similar results of carboxyl content for oxidized banana starch. The cleavage of glycosidic bonds during the oxidation process caused depolymerization of the starch molecules, which created conditions for the formation of carbonyl and carboxyl groups (Richardson and Gorton, 2003). Wang and Wang (2003) reported that the hydroxyl groups present in starch first transform into carbonyl groups and then into carboxyl groups during the oxidation of starch. The oxidizing agent, starch source, and reaction conditions also affect the differences in carboxyl contents of starches (Kuakpetoon and Wang, 2001).

3.3. Degree of cross-linking and molar substitution

The degree of cross-linking in cross-linked starch is determined by the values obtained from RVA analysis. In cross-linked starch, nearly 72.98% cross-linking was obtained which is very high due to the increased amount of phosphate salts. STMP/STPP increased the degree of cross-linking. Similar results for the degree of cross-linking were found by Koo et al. (2010) and Goswami et al. (2018) in their research on corn and white sorghum starch. The reduced swelling power of starch granules is due to the high degree of cross-linking, which also lowers peak viscosity (Xiao et al., 2012). These findings are also comparable with pasting and swelling results where lower peak viscosity and lower swelling values were obtained for cross-linked starch. Molar substitution (MS) of hydroxypropylated pigeon pea starch was 0.0009. The Molar substitution value of hydroxypropylated pigeon pea starch was within the range reported by FDA (7%, MS = 0.2) and this starch could be acceptable for food industry use (Dias et al., 1997).

3.4. Color evaluation

The color is a major criterion while modification of different types of starches. The color of native and modified pigeon pea starch samples was observed by Hunter color Flex colorimetric assay. The L* value of the native, oxidized, cross-linked and hydroxypropylated pigeon pea starch samples were 94.84, 94.87, 95.93, and 95.62 respectively. Native and oxidized pigeon pea starch showed a similar trend but cross-linked and hydroxypropylated starches have more whiteness than native starch. A small difference in the L* value of oxidized starch (94.87) and native starch (94.84) mentioned that 1.6 g of active chlorine concentration was not adequate to enhance starch whiteness. Similar findings were also reported by Sanchez-Rivera et al. (2005) for oxidized banana starch and they concluded that the L* value of starch will be increased with the increase in chlorine concentration. In an oxidation reaction process, a few pigments and proteins are oxidized before the glucose

units and thus produce starch brighter (Sanchez-Rivera et al., 2005).

3.5. Swelling power and solubility

The functional properties of native and chemically treated starches, viz swelling power and solubility are shown in Fig. 1a–b. The swelling power of starch depends upon amylose content and the water-holding capacity of starch molecules by hydrogen bonding (Liang and King, 2006). The swelling power of all starch samples showed increased with the rise in temperature and maximum swelling occurs between 75 °C and 95 °C. At 95 °C, the swelling power of oxidized and cross-linked starch was reduced in comparison to native starch, and the maximum reduction occurred in cross-linked starch (6.78%). Similar results of reduced swelling power have been reported by Kuakpetoon and Wang (2001) for oxidized common corn starch and Goswami et al. (2018) for white sorghum starch. The oxidation process resulted in the structural disintegration of starch granules and thus, decreased the swelling power of native starch. The decrease in swelling power of cross-linked starch could be due to the bridging of intermolecular bonds by phosphorous residual due to cross-linking (Chung et al., 2004). Hydroxypropylated modified starch showed higher swelling power (15.49%) in comparison to the native starch (12.49%). Increased swelling power in hydroxypropylated starch was also reported by Majzoobi et al. (2014) for wheat starch and Lawal (2011) for pigeon pea starch. During hydroxypropylation, alkaline treatment increased the pH which ionizes the hydroxyl groups in starch chains and disrupts the hydrogen bonds within the molecules increasing the granular swelling (Gray and BeMiller, 2005; Gunaratne and Corke, 2007). Native and chemically treated pigeon pea starch sample's solubility is shown in Fig. 1b. With the increase in temperature, the solubility of the starch samples was increased and maximum solubility occurs between 75 °C and 85 °C. At 95 °C, the solubility of hydroxypropylated and cross-linked starches decreased in comparison to the native starch, and the maximum reduction was observed for cross-linked starch (9.87%). Cross-linking treatment might have blocked the starch molecules from leaching out, making starch granules more compact and hence, less breakdown occurred during the gelatinization process resulting in lower solubility values (Xiao et al., 2012). An increase in solubility of starch after hydroxypropylation was due to the leached amylose content from the amorphous region (Lawal, 2011). Oxidized starch showed a higher solubility value (31.86%) in comparison to native starch (16.74%). The higher solubility of oxidized starch could be due to the weakening of starch granule structure and de-polymerization, thereby increasing the leaching of amylose from the starch granules (Ali and Hasnain, 2014). The higher solubility of oxidized starch as compared to other starch samples indicated that the granular structure of starch becomes weakened after oxidation.

3.6. Morphological properties

Native and chemically treated pigeon pea starch samples scanning electron micrographs (SEM) are shown in Fig. 2a–d. The starch morphology varies with its source and also depends upon chloroplast biochemistry (Singh et al., 2003). Thermal properties, starch digestibility, and physicochemical characteristics of starches are also affected by starch granule size and shape (Singh et al., 2007). SEM images of pigeon pea starch had large oval to elliptical-shaped granules with no evidence of any fissures, and no noticeable change was observed in the morphology of native and modified starch samples. Oval and kidney-shaped granules of legume starch were earlier reported by Hoover and Ratnayake (2002). The granule size of the native starch sample was in the range of 11.57–38.63 μm and modified starch samples were in the range of 18.85–22.58 μm. The larger size of the native starch granule might be responsible for high PV, HPV, CPV, and SB in this study (Rincon-Londono et al., 2016). Gelatinization temperature, pasting characteristics, enzyme susceptibility, crystallinity, and starch
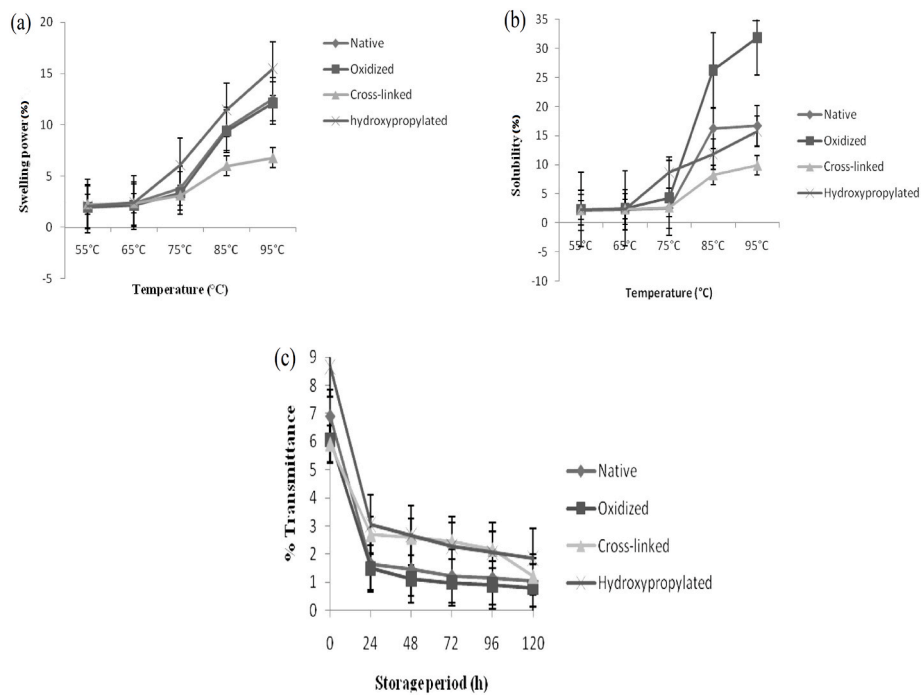
Fig. 1. (a) Swelling power and (b) solubility of native and chemically treated pigeon pea starch samples; (c) % transmittance of native and chemically treated pigeon pea starch samples.

solubility, and all these properties are affected by starch granule size (Lindeboom et al., 2004). In oxidized starch, no noticeable difference was observed between the appearance of the native and the oxidized starch granule. This showed that the levels of hypochlorite (1.6 g/100 g active chlorine) used in oxidation did not cause any significant change in the morphology of starch granules. Similar results regarding changes in granule morphology have been reported previously (Kuakpetoon and Wang, 2001; Lawal, 2004; Sandhu et al., 2008). Other authors, however, have demonstrated that oxidation can affect the granule with the presence of pores on the surface (Kuakpetoon and Wang, 2008). Spier et al. (2013) modified corn starch and noticed that oxidation in conjunction with an alkaline treatment caused holes in the granule surface. Vanier et al. (2012) found some changes in the external structure of bean starch and its surface appears to be rougher after 1.5% active chlorine oxidation. Similarly, the cross-linked and hydroxypropylated starch external surface appeared to be smooth with no noticeable pores will appear which indicates that the orderly crystalline structure was not destroyed after these modifications. Although, Koo et al. (2010) and Carmona-Garcia et al. (2009) observed slight changes, black zones on the surface along the longitudinal axis, in corn and banana starch granules when cross-linked with STMP/STPP.

3.7. Fourier transform-infrared spectroscopy (FTIR)

FT-IR spectroscopy has been used as a quantitative tool to describe the organization and structural variations in the starch molecule. Data obtained from FTIR is related to the short-range order in the starch molecule (Bello-Perez et al., 2005). The FT-IR spectra of native and chemically treated pigeon pea starch samples are shown in Fig. 2e and it could be observed that no structural changes were observed between native and modified starch samples after modification. Native and modified starch samples have similar characteristic peaks but a change in the absorbance ratio 1047/1022 cm^{-1} of chemically treated pigeon pea starch samples was observed and the absorption intensity of all chemically treated starches was smaller than the native starch. The modification methods had a considerable effect on the IR ratio of pigeon pea starch and the 1047/1022 cm^{-1} ratio was 1.09, 1.08, 1.04, 1.02 for

native, oxidized, hydroxypropylated, and cross-linked starch respectively. The decrease in the IR ratio of chemically treated starch samples might be due to the separating and unraveling of double helices forming the crystalline array. The decrease in the IR ratio in chemically modified starches was also reported by Halal et al. (2015). In the spectra of native pigeon pea starch, there were absorbance bands at 1148, 1076, and 991 cm^{-1} attributed to C–O bond stretching. Significant differences were observed in native and chemically treated pigeon pea starch samples with intensity bands appearing between 3255 and 2921 cm^{-1} and which shows the presence of O–H and C–H bond stretching. An extremely broadband due to hydrogen-bonded hydroxyl groups appeared at 3288 cm^{-1} but was slightly reduced after modification. The absorption peak at 1632 cm^{-1} is due to the shaking of water molecules occupying the non-crystalline region of the starch molecule (Kizil et al., 2002). The spectra showed that native and oxidized starch had near similar absorption intensity and the same result was also observed by Liu et al. (2014) for oxidized maize starch.

3.8. Paste clarity

Paste clarity reflects the transmission of the light through the starch paste. The results of paste clarity or % transmittance of native and chemically treated pigeon pea starches stored for 5 days at 4 °C differed significantly ($p < 0.05$) and are presented in Fig. 1c. In all starch samples, paste clarity was significantly reduced ($p < 0.05$) with a rise in storage time. The maximum reduction in paste clarity was observed after 24 h of cold storage and after 120 h. Oxidized starch had minimum paste clarity (0.81%), while the maximum was observed for hydroxypropylated starch (1.86%). Similar findings were also reported by Xiao et al. (2012) for cross-linked rice starch and Khan et al. (2014) for oxidized potato starch. Cross-linking treatment gives strength to the swollen starch granules which results in a decrease in light transmittance. The reduction in paste clarity in all starch samples during storage at cold temperatures was due to the molecular restructuring of solubilized starch chains (Goswami et al., 2018).
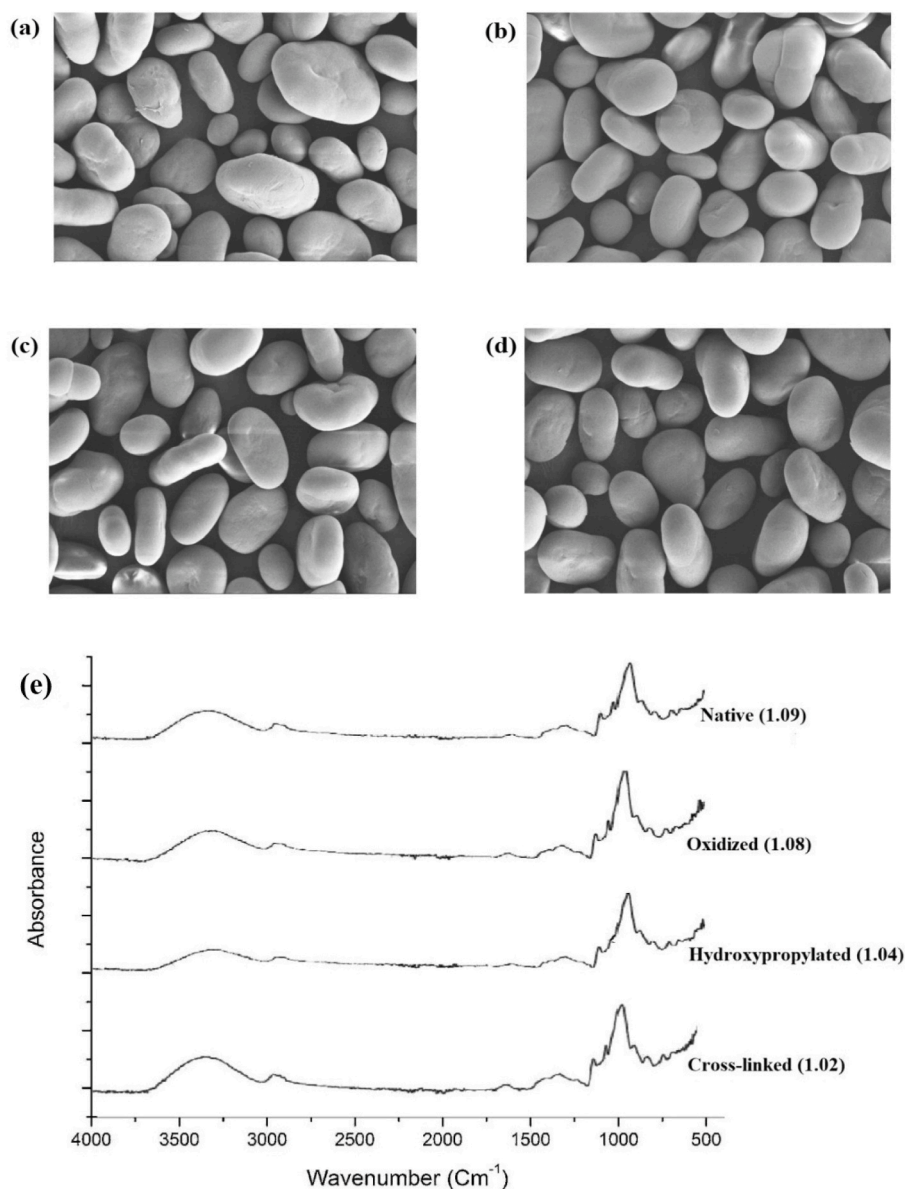
Fig. 2. Scanning electron micrographs of native and chemically treated pigeon pea starch samples under magnification 2.50 KX (a) native; (b) oxidized; (c) cross-linked; (d) hydroxypropylated; (e) Fourier transform infrared spectroscopy ratios of 1047 cm^{-1}/1022 cm^{-1} for native and chemically treated pigeon pea starch samples.

3.9. Starch gel hardness

A texture analyzer is used to measure the hardness of starch gel (Yu et al., 2016). An increase in gel hardness in starch mainly results from the retrogradation which occurs in amylose and amylopectin components (Karim et al., 2008), and the inhibition of amylose chain interactions reducing the formation of junction zones results in a decrease in starch gel hardness (Liu et al., 1999). Significant differences ($p <$ 0.05) in the gel hardness of native and chemically treated pigeon pea starch samples were observed. Gel hardness showed a decreasing trend after modification in comparison to the native starch. Gel hardness in native and chemically treated pigeon pea starch samples varied from 200.52 to 712.51 g. The modification affects the hardness value of gels and the lowest hardness value (200.52 g) was observed for cross-linked while the native starch gel (712.51 g) had the highest value. Cross-linking decreased the amount of amylose leaching resulting in a decrease in the amount of amylose in the continuous network and thus, forming a weak gel structure (Gunaratne and Corke, 2007). These results

are also comparable with the lower cold paste viscosity values of cross-linked starch. A similar result for the reduction in gel hardness for hydroxypropylated starch was also observed by Liu et al. (1999) and they stated that the hydroxypropyl groups suppress the starch chain association and thus the solidification of starch gel can be effectively retarded by using hydroxypropylation.

3.10. RVA characteristics

The pasting characteristics are an essential parameter to determine the use of starch in different food products. The pasting characteristics of native and chemically treated pigeon pea starches obtained by RVA are presented in Table 1 and Fig. 3. The pasting temperature of native and chemically treated pigeon pea starch samples varied from 70.81 to 84.60 °C. After chemical modification, the pasting temperature in all modified starches was increased and the highest increase occurred in cross-linked starch. This behavior is different from the results of Spier et al. (2013). They found that the pasting temperatures of sodium

Table 1
RVA viscosity characteristics of native and chemically treated pigeon pea starch samples.

Starch samples	PV (cP)	HPV (cP)	BD (cP)	CPV (cP)	SB (cP)	Pasting Temp (°C)	Peak Time (Min)	Stability ratio	Setback ratio
Native	4586 ± 97.63^d	3449 ± 75.79^c	1183 ± 23.71^c	6153 ± 61.55^d	3060 ± 103.16^d	70.81 ± 17.94^a	4.87 ± 0.01^b	0.75	1.78
Oxidized	4135 ± 146.11^b	3308 ± 96.51^c	854 ± 41.32^b	5043 ± 85.04^b	1675 ± 63.12^b	82.36 ± 0.05^c	5.08 ± 0.01^c	0.80	1.53
Cross-linked	1209 ± 48.26^a	1147 ± 43.66^a	62 ± 5.56^a	2020 ± 107.56^a	793 ± 45.09^a	84.60 ± 0.17^d	7.00 ± 0.00^d	0.94	1.76
Hydroxypropylated	4266 ± 98.43^c	2909 ± 97.00^b	1353 ± 56.32^d	5385 ± 149.00^c	2413 ± 29.30^c	80.71 ± 0.02^b	4.80 ± 0.00^a	0.68	1.85

Values are means of triplicate analyses with standard deviation.
Value presents in the same column with different superscripts were significantly different (p < 0.05).
PV Peak viscosity, BD Breakdown, HPV Hot paste viscosity, SB Setback, CPV Cold paste viscosity.
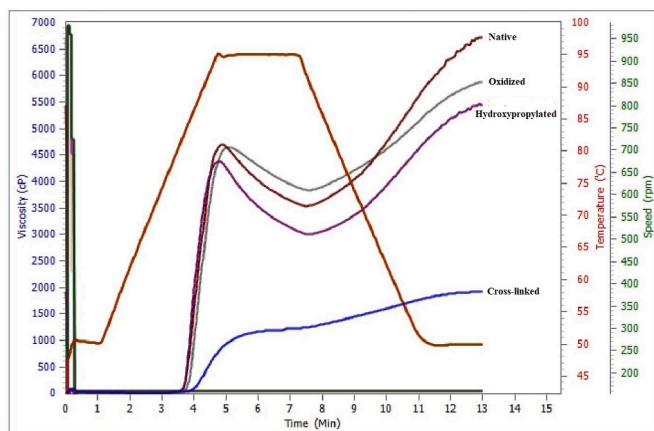Stability ratio (HPV/PV) and Set back ratio (CPV/HPV).

Fig. 3. Pasting characteristics of native and chemically treated pigeon pea starch samples.

hypochlorite-oxidized corn starch were lower than that of the native corn starch. The peak time of native and chemically treated pigeon pea starches varied from 4.80 to 7.00 min, being lowest for native starch and highest for cross-linked starch. Starches having lower peak times are easy to cook and starches having higher peak times need more time for cooking (Yamani et al., 2013). The lowest value of peak viscosity was obtained for cross-linked starch (1222 cP), while the highest value was obtained for native starch (4524 cP). The high peak viscosity of native starch gel paste might be due to the formation of the firm hydrogel as compared to other modified starch paste. A reduction in peak, cold paste, hot paste, and setback viscosity was observed for all chemically treated starches and cross-linked starch exhibited the lowest values for all pasting parameters except pasting temperature and peak time. The reduction in the pasting viscosities of modified starches could be ascribed to the fact that reorganization occurred within the starch granules after chemical modification, which restricted the swelling capacity of starch and less amount of amylose leached into the medium to increase its viscosity. The breakdown viscosity of starch was used to determine its fragility. (Vanier et al., 2012). The observed breakdown viscosity values ranged from 62 to 1353 cP, with native starch having the lowest and hydroxypropylated starch having the highest. A high breakdown viscosity is closely linked to a higher swelling ability of the starch granules and a decrease in the shear resistance of the starch paste during the heating process. As a result, the lower the breakdown, the better the thermal stability and shear resistance of the starch paste (Chan et al., 2010). Kuakpetoon and Wang (2006) also reported a decrease in the breakdown viscosity of corn starch. When compared to native starch, the setback (SB) viscosity in all chemically treated starch samples was reduced. Lower setback viscosities indicated a lower tendency of the

starch granules to retrograde (Sandhu et al., 2008). When a shear force is applied to a starch paste, the stability ratio indicates resistance to breakdown. After modification, a paste made from cross-linked starch was more stable to shear (stability ratio 0.94) than a paste made from hydroxypropylated starch (stability ratio 0.68). An increase in the stability ratio might be attributed to an improved organization within the starch granules after modification, which resulted in restricted swelling and low mechanical destruction. The setback ratio is directly related to the tendency of starch to retrograde after gelatinization. The hydroxypropylated starch paste had the highest (1.85) setback ratio, while oxidized starch had the lowest (1.53).

3.11. Rheological properties

3.11.1. Steady shear properties

The rheological properties of starch paste are affected by many factors such as starch source, the concentration of amylose/amylopectin ratio, pasting temperature, the concentration of starch, and pH (Choi and Yoo, 2009). The experimental data of shear stress (σ) versus shear rate (γ) for native and modified starches are presented in Fig. 4a–b and Table 2A. The steady shear properties determination coefficients are best explained with a Herschel-Bulkley model ($R^2 = 0.998$ to 0.999). In starch rheology 'n' value is used to define the fluid and semi-fluid nature of starch pastes where n < 1 shows the shear thinning behavior while n > 1 indicates the shear thickening behavior of starch pastes. The Herschel-Bulkley model revealed that the n value for all starch pastes varied from 0.45 to 0.85, the minimum and maximum values were observed for cross-linked and native starch, respectively. A higher shear rate is responsible for the breakage of inter and intramolecular binding of starch micelles resulting in the shear-thinning behavior of starch pastes (Bhandari et al., 2002). The consistency index (K) tells about the structural strength and values of K for all starch samples varied from 1.18 to 8.49 ($Pa.s^n$) and cross-linked starch had the highest K value. The minimum force required to start a flow of starch paste is called σo (yield stress). The yield stress values ranged from 5.95 to 86.35 Pa, being the highest for hydroxypropylated starch, indicating that it required a higher force to initiate the flow. Cross-linked starch pastes showed lesser values of K and σo than native starch and the reason may be the cross-linked starch granules have low solubility and swelling power (Kim and Yoo 2010). These results were also evident in the low solubility of cross-linked starch (Fig. 1b).

3.11.2. Dynamic shear properties

In a study of dynamic rheological properties of starch, storage modulus (G′) and loss modulus (G″) defines the structure of a starch gel. The storage modulus (G′) is a measure of the elastic response of the material and measured stored energy in a material per deformation cycle while the loss modulus (G″) is a measure of the viscous response of the material and measured the energy dissipated from the material per
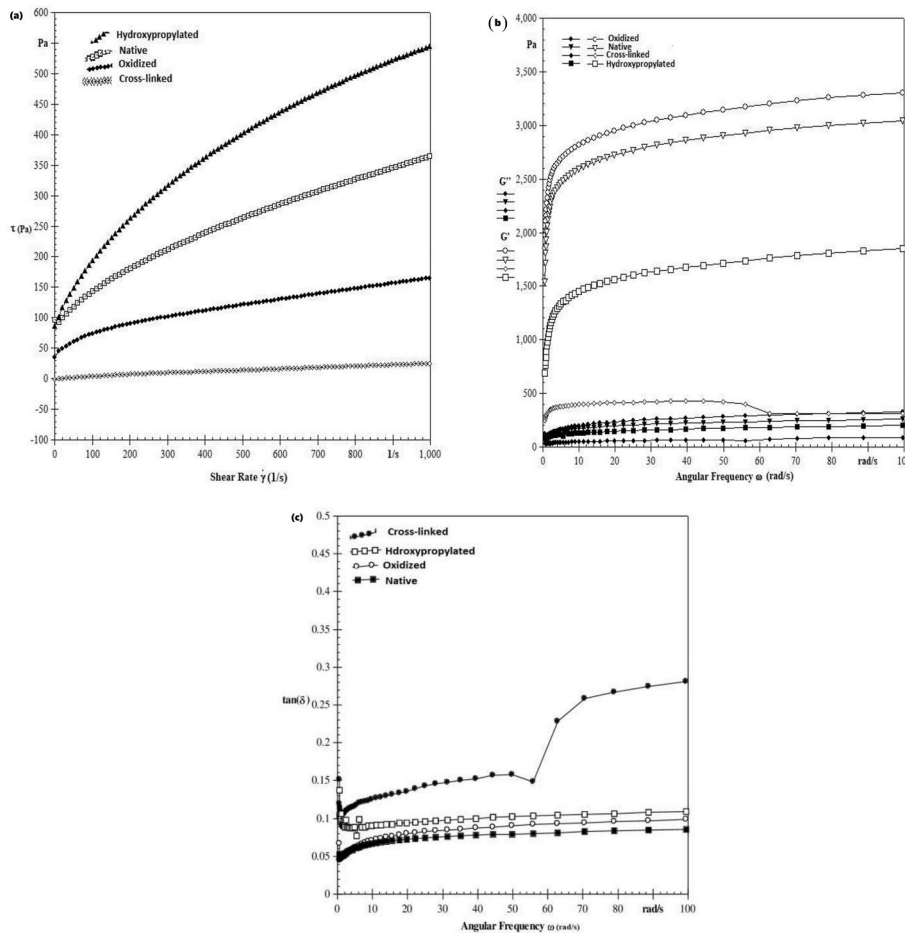
Fig. 4. Dynamic rheological properties; and (a) Flow properties of native and chemically treated pigeon pea starch pastes (10% w/w); (b) Dynamic mechanical spectra of G′ and G″ of native and chemically treated pigeon pea starch pastes; (c) Angular frequency dependence of tan δ at 25 °C.

Table 2A
Flow behavior parameters of native and chemically treated pigeon pea starch samples.

Starch samples	σ ο (Pa)	K (Pa.s)	n	R^2
Native	84.15 ± 0.31[c]	1.16 ± 0.01[a]	0.85 ± 0.00[d]	0.998
Oxidized	35.54 ± 0.59[b]	2.12 ± 0.02[b]	0.63 ± 0.01[b]	0.999
Cross-linked	5.73 ± 0.22[a]	8.50 ± 0.03[d]	0.45 ± 0.00[a]	0.999
Hydroxypropylated	86.65 ± 0.39[d]	2.36 ± 0.16[c]	0.83 ± 0.00[c]	0.998

Values are means of triplicate analyses with standard deviation.
Value presents in the same column with different superscripts were significantly different (p < 0.05).

deformation cycle. The storage modulus (G′) and loss modulus and tan δ as a function of frequency observed at 25 °C are shown in Fig. 4c and data is presented in Table 2B. A rise in the value of G′ and G″ starch paste

Table 2B
Rheological properties of native and chemically treated pigeon pea starch samples during the frequency sweep (0–100 rad/s).

Starch samples	G′ (Pa) (Storage modulus)	G″ (Pa) (Loss modulus)	tan (δ)
Native	2517 ± 11.23[c]	157 ± 2.08[c]	0.05 ± 0.00[a]
Oxidized	1364 ± 3.60[b]	135 ± 2.00[b]	0.08 ± 0.00[b]
Cross-linked	384 ± 4.50[a]	46 ± 2.00[a]	0.12 ± 0.01[c]
Hydroxypropylated	2730 ± 4.35[d]	176 ± 2.51[d]	0.06 ± 0.01[a]

Values are means of triplicate analyses with standard deviation.
Value presents in the same column with different superscripts were significantly different (p < 0.05).

was seen with an increase in the angular frequency. All the starch pastes had a higher value for G′ as compared to G″, which shows that starch pastes had visco-elastic behaviour. G′ and G″ showed no cross-over during the analyzed frequency range (0.1–100 rad/s) showing the stability of starch paste during this range. This type of behavior was also reported for cross-linked and hydroxypropylated wheat starch (Yousefi and Razavi, 2015; Morikawa and Nishinari, 2000; Lee and Yoo, 2011). The loss tangent (tan ∂ = G″/G′) represents the dynamic elastic behavior of gel (Toker et al., 2013). The low tan ϱ value shows the elastic behavior of paste and the high value shows the viscous behavior of paste. The value of tan δ (G″/G′) of starch pastes was <1 showing more elastic compared to viscous behavior. Studying rheological properties is very important to regulate the production processes, transfer of sauces with pumps, and packaging of liquid food products in bottles. Rheology studies also provide information for product development, quality control, process design, and sensory evaluation (Kulicke et al., 1996; Yousefi and Razavi, 2015).

4. Conclusion

Chemical modifications had a significant (*p*< 0.05) effect on the physicochemical, pasting, and rheological properties of pigeon pea starch. Paste clarity, gel hardness, swelling power, and solubility of modified starches were significantly decreased after the chemical modification. All modified starches had lower peak viscosity, cold paste, hot paste, and setback viscosity indicating less swelling of starch granules and lower rates of starch retrogradation. The stability ratio was increased after the starch modification and the paste from cross-linked

starch was more stable to shear whereas; the paste from hydroxypropylated starch was least stable to shear. SEM analysis showed no significant changes after chemical modifications and this pattern is related to a low degree modification obtained with all reagents used for modification. In the frequency sweep test G′ value showed a higher value as compared to G″ value at the observed frequency range of 0.1–100 rad/s which shows that starch pastes had a visco-elastic behavior. tan **δ** value was observed as less than 1 for all starches indicating their elastic behavior than viscous. In the overall investigation, the cross-linked starch exhibited a more stable structure than the other modified starches; thus, this modification is better than the others since it showed the highest thermal stability.

CRediT authorship contribution statement

Prixit Guleria: Writing – original draft, Methodology, Conceptualization, Visualization, Data curation. **Baljeet Singh Yadav:** Writing – review & editing, and, Supervision.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Data availability

The data that has been used is confidential.

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References

Adebowale, K.O., Afolabi, T.A., Lawal, O.S., 2002. Isolation, chemical modification and physicochemical characterization of Bambarra groundnut (*Voandzeia subterranean*) starch and flour. Food Chem. 78, 305–311.

Ali, T.M., Hasnain, A., 2014. Morphological, physiochemical, and pasting properties of modified white sorghum (*Sorghum Bicolor*) starch. Int. J. Food Prop. 17, 523–535.

Aoac, 1995. Official Methods of Analysis, sixteenth ed. Association of Official Analytical Chemists, Washington, DC.

Bello-Perez, L.A., Francisco, A.D., Agama-Acevedo, E., Gutierrez-Meraz, F., Garcia-Suarez, F.J.L., 2005. Morphological and molecular studies of banana starch. Food Sci. Technol. Int. 11, 367–372.

Bhandari, P.N., Singhal, R.S., Kale, D.D., 2002. Effect of succinylation on the rheological profile of starch pastes. Carbohydr. Polym. 47, 365–371.

Carmona-Garcia, R., Sanchez-Rivera, M.M., Mendez-Montealvo, G., Garza- Montoya, B., Bello-Perez, L.A., 2009. Effect of the cross-linked reagent type on some morphological, physicochemical and functional characteristics of banana starch (*Musa paradisiaca*). Carbohydr. Polym. 76, 117–122.

Chan, H.T., Bhat, R., Karim, A.A., 2010. Effects of sodium dodecyl sulphate and sonication treatment on physicochemical properties of starch. Food Chem. 12, 703–709.

Chattopadhyay, S., Singhal, R.S., bKulkarni, P.R., 1997. Optimisation of conditions of synthesis of oxidized starch from corn and amaranth for use in film-forming applications. Carbohydr. Polym. 34, 203–212.

Choi, H.M., Yoo, B., 2009. Steady and dynamic shear rheology of sweet potato starch–xanthan gum mixtures. Food Chem. 116, 638–643.

Chung, H.J., Liu, Q., Donner, E., Hoover, R., Warkentin, T.D., Vandenberg, B., 2008. Composition, molecular structure, properties, and in vitro digestibility of starches from newly released Canadian pulse cultivars. Cereal Chem. 85, 471–479.

Chung, H.J., Woo, K.S., Lim, S.T., 2004. Glass transition and enthalpy relaxation of cross-linked corn starches. Carbohydr. Polym. 55, 9–15.

Dias, F.F., Tekchandani, H.K., Mehta, D., 1997. Modified starches and their use by food industries. Food Chem. 64, 361–375.

Emefiene, M.E., Joshua, V.I., Nwadike, C., Yaroson, A.Y., Zwalnan, N.D.E., 2014. Profitability analysis of pigeon pea (*Cajanus cajan*) production in Riyom LGA of Plateau state. Int. Lett. Nat. Sci. 18, 73–88.

Esan, V., Ojemola, O., 2018. Evaluation of production systems, traditional knowledge of pigeon pea (*Cajanus cajan*) and risks of extinction of pigeon pea, jack bean (*Canavalia ensiformis*) and lubia bean (*Lablab purpureus*) in some parts of South West Nigeria. J. Exp. Agric. Int. 21, 1–11.

Forssell, P., Hamunen, A., Autio, K., Suortti, P., Poutanen, K., 1995. Hypochlorite oxidation of barley and potato starch. Starch Staerke 47, 371–377.

Fonseca, L.M., Gonçalves, J.R., Mello El Halal, S.L., Pinto, V.Z., Dias, A.R.G., Jacques, A.C., Zavareze, E.D.R., 2015. Oxidation of potato starch with different sodium hypochlorite concentrations and its effect on biodegradable films. LWT–Food Sci. Technol. 60, 714–720.

Garcia, E.A., Feria, J.S., Villalobos, J.R.R., Gonzalez, F.R., Perez, A.J., Huicochea, E.F., 2014. Properties of edible films based on oxidized starch and zein. Int. J. Polym. Sci. 9. Article ID 292404.

Goswami, K., Yadav, R.B., Yadav, B.S., Yadav, R.L., 2018. Physico-chemical, textural and crystallinity properties of oxidized, cross-linked and dual-modified white sorghum starch. Int. Food Res. J. 25, 2104–2111.

Gray, J.A., BeMiller, J.N., 2005. Influence of reaction conditions on the location of reactions in waxy maize starch granules reacted with a propylene oxide analog at low substitution levels. Carbohydr. Polym. 60, 147–162.

Gunaratne, A., Corke, H., 2007. Functional properties of hydroxypropylated, cross-linked, and hydroxypropylated cross-linked tuber and root starches. Cereal Chem. 84, 30–37.

Gunasekaran, S., Mehmet, M.A.K., 2000. Dynamic oscillatory shear testing of foods-selected applications. Trends Food Sci. Technol. 11, 115–127.

Halal, S.L.M., Colussi, R., Pinto, V.Z., Bartz, J., Radunz, M., Carreno, N.L.V., Dias, A.R.G., Zavareze, E.R., 2015. Structure, morphology, and functionality of acetylated and oxidized barley starches. Food Chem. 168, 247–256.

Hjermstad, E.T., 1971. Highly hydroxypropylated granule potato starch. U.S. Patent 3 (577), 407.

Hood, L.F., Mercier, C., 1978. Molecular structure of unmodified and chemically modified manioc starches. Carbohydr. Res. 61, 53–66.

Hoover, R., Ratnayake, W.S., 2001. Determination of total amylose content of starch. Curr. Protoc. Food Anal. Chem. (1), E2, 3.1-E2. 3.5.

Hoover, R., Ratnayake, W.S., 2002. Starch characteristics of black bean, chickpea, lentil, navy bean and pinto bean cultivars grown in Canada. Food Chem. 78, 489–498.

Karim, A.A., Zaki, N., Chen, F.K., Phuah, Y.P., 2008. Pasting and retrogradation properties of alkali-treated sago (*Metroxylan sago*) starch. Food Hydrocolloids 22, 1044–1053.

Khan, K.H., Ali, T.M., Hasnain, A., 2014. Effect of chemical modifications on the functional and rheological properties of potato (*Solanum tuberosum*) starches. J. Anim. Plant Sci. 24, 550–555.

Kim, B.Y., Yoo, B., 2010. Effects of cross-linking on the rheological and thermal properties of sweet potato starch. Starch Staerke 62, 577–583.

Kizil, R., Irudayaraj, J., Seetharaman, K., 2002. Characterization of irradiated starches by using FT-Raman and FTIR-Spectroscopy. J. Agric. Food Chem. 50, 3912, 3818.

Koo, S.H., Lee, K.Y., Lee, H.G., 2010. Effect of cross-linking on the physicochemical and physiological properties of corn starch. Food Hydrocolloids 24, 619–625.

Koocheki, A., Razavi, S.M., 2009. Effect of concentration and temperature on flow properties of Alyssum homolocarpum seed gum solutions: assessment of time dependency and thixotropy. Food Biophys. 4, 353–364.

Kuakpetoon, D., Wang, Y.J., 2001. Characterization of different starches oxidized by hypochlorite. Starch Staerke 53, 211–218.

Kuakpetoon, D., Wang, Y.J., 2006. Structural characteristics and physicochemical properties of oxidized corn starches varying in amylose content. Carbohydr. Res. 341, 1896–1915.

Kuakpetoon, D.S., Wang, Y.J., 2008. Locations of hypochlorite oxidation in corn starches varying in amylose content. Carbohydr. Res. 343, 90–100.

Kulicke, W.M., Kull, A.H., Kull, W., Thielking, H., Engelhardt, J., Pannek, J.B., 1996. Characterization of aqueous carboxymethylcellulose solutions in terms of their molecular structure and its influence on rheological behavior. Polymer 37, 2723–2731.

Lauzon, R.D., Shiraishi, K., Yamazaki, M., Sawayama, S., Sugiyama, N., Kawabata, A., 1995. Physicochemical properties of cocoyam starch. Food Hydrocolloids 9, 77–81.

Lawal, O.S., 2004. Composition, physicochemical properties and retrogradation characteristics of native, oxidized, acetylated and acid-thinned new cocoyam (*Xanthosoma sagittifolium)* starch. Food Chem. 87, 205–218.

Lawal, O.S., Storz, J., Storz, H., Lohmann, D., Lechner, M.D., Kulicke, W.M., 2009. Hydrogels based on carboxymethyl cassava starch cross-linked with di-or polyfunctional carboxylic acids: synthesis, water absorbent behaviour and rheological characterizations. Eur. Polym. J. 45, 3399–3408.

Lawal, O.S., 2011. Hydroxypropylation of pigeon pea (Cajanus cajan) starch: preparation, functional characterizations and enzymatic digestibility. LWT–Food Sci. Technol. 44, 771–778.

Lee, H.L., Yoo, B., 2011. Effect of hydroxypropylation on physical and rheological properties of sweet potato starch. LWT–Food Sci. Technol. 44, 765–770.

Liang, X., King, J.M., 2006. Pasting and crystalline property differences of commercial and isolated rice starch with added amino acids. J. Food Sci. 68, 832–838.

Lindeboom, N., Chang, P.R., Tyler, R.T., 2004. Analytical, biochemical and physicochemical aspects of starch granule size, with emphasis on small granule starches: a review. Starch Staerke 56, 88–99.

Liu, H., Ramsden, L., Corke, H., 1999. Physical properties and enzymatic digestibility of hydroxypropylated ae, wx, and normal maize starch. Carbohydr. Polym. 40, 175–182.

Liu, J., Wang, B., Lin, L., Zhang, J., Liu, W., Xie, J., Ding, Y., 2014. Functional, physicochemical properties and structure of cross-linked oxidized maize starch. Food Hydrocolloids 36, 45–52.

Majzoobi, M., Saberi, B., Farahnaky, A., Mesbahi, G., 2014. Comparison of physicochemical and gel characteristics of hydroxypropylated oat and wheat starches. Int. J. Food Eng. 10, 657–667.

Morikawa, K., Nishinari, N., 2000. Effects of concentration dependence of retrogradation behaviour of dispersions for native and chemically modified potato starch. Food Hydrocolloids 14, 395–401.

Moore, C.O., Tuschhoff, J.V., Hastings, C.W., Schanefelt, R.V., 1984. Application of starches in foods. In: Starch: Chemistry and Technology, 2nd. Academic Press, Orlando, Florida, pp. 579–592.

Naknaen, P., Tobkaew, W., Chaichaleom, S., 2017. Properties of jackfruit seed starch oxidized with different levels of sodium hypochlorite. Int. J. Food Prop. 20, 979–996.

Narina, S.S., Xu, Y., Hamama, A.A., Phatak, S.C., Bhardwaj, H.L., 2012. Effect of cultivar and planting time on resistant starch accumulation in pigeon pea grown in Virginia. ISRN Agron. Article ID 576471, 4 pages.

Pelissari, F.M., Yamashita, F., Garcia, M.A., Martino, M.N., Zaritzky, N.E., Grossmann, M.V.E., 2012. Constrained mixture design applied to the development of cassava starch-chitosan blown films. J. Food Eng. 108, 262–267.

Richardson, S., Gorton, L., 2003. Characterisation of the substituent distribution in starch and cellulose derivatives. Anal. Chim. Acta 497, 27–65.

Rincon-Londono, N., Vega-Rojas, L.J., Contreras-Padilla, M., Acosta-Osorio, A.A., Rodríguez-García, M.E., 2016. Analysis of the pasting profile in corn starch: structural, morphological, and thermal transformations, part I. Int. J. Biol. Macromol. 91, 106–114.

Sandhu, K.S., Kaur, M., Singh, N., Lima, S.T., 2008. A comparison of native and oxidized normal and waxy corn starches: physicochemical, thermal, morphological and pasting properties. LWT–Food Sci. Technol. 41, 1000–1010.

Sanchez-Rivera, M.M., Garcia-Suarez, F.J., Velazquez, D.V.M., Gutierrez- Meraz, F., Bello-Perez, L.A., 2005. Partial characterization of banana starches oxidized by different levels of sodium hypochlorite. Carbohydr. Polym. 62, 50–56.

Sharma, P., Kaur, M., Sandhu, K.S., Nain, V., Janghu, S., 2021. Physicochemical and rheological properties of cross-linked litchi kernel starch and its application in development of bio-films. Starch Staerke 73, 2100049.

Singh, J., Kaur, L., McCarthy, O.J., 2007. Factors influencing the physico-chemical, morphological, thermal and rheological properties of some chemically modified starches for food applications. A review. Food Hydrocolloids 21, 1–22.

Singh, N., Singh, J., Kaur, L., Sodhi, N.S., Gill, B.S., 2003. Morphological, thermal and rheological properties of starches from different botanical sources. Food Chem. 81, 219–231.

Smith, R.J., 1967. Characterization and analysis of starches. In: Whistler, R.L. (Ed.), Science of Chemistry and Technology. Academic Press, New York, pp. 620–625.

Spier, F., Zavareze, E.R., Silva, R.M., Elias, M.C., Dias, A.R.G., 2013. Effect of alkali and oxidative treatments on the physicochemical, pasting, thermal and morphological properties of corn starch. J. Sci. Food Agric. 93, 2331–2337.

Toker, O.S., Karaman, S., Yuksel, F., Dogan, M., Kayacier, A., Yilmaz, M.T., 2013. Temperature dependency of steady, dynamic and creep-recovery rheological properties of ice cream mix. Food Bioprocess Technol. 6, 2974–2985.

Vanier, N.L., Zavareze, E.R., Pinto, V.Z., Klein, B., Botelho, F.T., Dias, A.R.G., Elias, M.C., 2012. Physicochemical, crystallinity, pasting and morphological properties of bean starch oxidized by different concentrations of sodium hypochlorite. Food Chem. 131, 1255–1262.

Wang, Y., Wang, L., 2003. Physicochemical properties of common and waxy corn starches oxidized by different levels of sodium hypochlorite. Carbohydr. Polym. 52, 207–217.

Wojeicchowski, J.P., Siqueira, G.L.A., Lacerda, L.G., Schnitzler, E., Demiate, I.M., 2018. Physicochemical, structural and thermal properties of oxidized, acetylated and dual-modified common bean (*Phaseolus vulgaris L.*) starch. Food Sci. Technol. 38, 318–327.

Xiao, H.X., Lin, Q.L., Liu, G.Q., Yu, F.X., 2012. A comparative study of the characteristics of cross-linked, oxidized and dual-modified rice starches. Molecules 17, 10946–10957.

Yadav, B.S., Guleria, P., Yadav, R.B., 2013. Hydrothermal modification of Indian water chestnut starch: influence of heat-moisture treatment and annealing on the physicochemical, gelatinization and pasting characteristics. LWT–Food Sci. Technol. 53, 211–217.

Yamani, B.V., Sanabria, G.G.R., Filho, F.F., 2013. The physical, chemical and functional characterization of starches from Andean tubers: oca *(Oxalis tuberosa Molina), olluco (Ullucus tuberosus Caldas) and mashua (Tropaeolum tuberosum Ruiz & Pavón)* Braz. J. Pharmacol. Sci. 49, 453–464.

Yousefi, A.R., Razavi, M.A., 2015. Dynamic rheological properties of wheat starch gels as affected by chemical modification and concentration. Starch Staerke 66, 1–10.

Yu, S., Liu, J., Yang, Y., Ren, J., Zheng, X., Kopparapu, N.K., 2016. Effects of amylose content on the physicochemical properties of Chinese chestnut starch. Starch Staerke 68, 112–118.