

Isolation And Characterization Of Starch From Quinoa (*Chenopodium Quinoa Willd.*)

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Abstract: *The physicochemical, functional and structural properties of starch isolated from lysine-rich high protein Chenopodium quinoa grains were studied. Quinoa starch had lower amylose content than other cereal starches. Solubility and swelling power of the quinoa starch increased with increasing temperature. The SEM observation revealed polygonal shape of starch granules. XRD showed a typical A- type diffraction pattern with high crystallinity (38.4 %) of quinoa starch. Thermal properties were studied by DSC viz. gelatinization temperatures $T_0(50.36^\circ c)$, $T_p(96.49^\circ c)$, $T_c(146.00^\circ c)$ and the enthalpies of gelatinization (ΔH_{gel}) (275.36 J/g). The pasting properties were studied by RVA showed peak viscosity (3593.00 cP), trough viscosity (3336.00 cP), breakdown viscosity (257.00 cP), set back viscosity (668.00 cP) and final viscosity (4004.00 cP) of the quinoa starch.*

Keywords: *Chenopodium quinoa, thermal properties, pasting properties, starch.*

I. INTRODUCTION

Quinoa (*Chenopodium quinoa* Willd.) is a pseudocereal native to the Andean regions of South America. It has been cultivated in the Andes since at least 3,000 B.C. (Wood, 1985). Peru is the largest producer of quinoa in the world, followed by Bolivia, the United States, Canada, and Ecuador. In the Quechua language of the Incas, quinoa is the *chisya mama* or “mother grain”.

In recent years, there has been renewed interest in quinoa because of its unique and interesting properties. Its starch exists as very small granules and is reported to be low in amylose (Lorenz, 1990). Its protein content and quality, with balanced amino acid composition similar to that of casein, are high compared to those of true cereals. But, beyond their nutritional function of supplying nutrients, quinoa provides compounds with promoting health properties such as phenolic acids, flavonoids and phytosterols (Abugoch James, 2009). In addition, quinoa is rarely allergenic because of the absence of gluten (Berti et al., 2004). Hence, it could be used in foods designed to reduce allergies in sensitive individuals, such as people with celiac disease, and it seems ideal for foods such as

infant formulae (Coulter and Lorenz 1990; Javaid 1997; Morita et al., 2001). The Food and Agriculture Organization claim quinoa is a crop that is destined to provide food security in the future due to its strong nutritional profile and the ability of new strains to grow in regions such as Africa, North American, and Asia (Jacobsen, 2000).

Quinoa produces high-protein grains under environmental extreme conditions that makes it important for the diversification of future agricultural systems. Currently, nutritionists, agronomists, and the food industry are evaluating quinoa in terms of genotype improvement, agronomics and processing to encourage its further cultivation as a crop in other parts of the world. This gives it the potential to become an important industrial and food crop that could contribute to diminish the shortage of provisions in developing countries (Bhargava et al., 2006). Nowadays, quinoa production is in a process of expansion into different geographic areas of the world due to its extraordinary adaptability. The genetic variability of quinoa is believed to be high, with cultivars being adapted to growth from sea level to 4000 m above sea level, from cold, highland climates to subtropical conditions. Quinoa is frost and drought resistant and can grow even in

regions where the annual rainfall is in the range of 200-400 mm (Valencia-Chamorro, 2003).

The United Nations Food and Agriculture Organization (FAO) (Johnson and Croissant, 1985) states its quality is "equal to the protein of whole dried milk." The protein component has an amino acid profile that very closely parallels the ideal protein standard sanctified by the FAO. The major component in quinoa consists of carbohydrates, and varies from 67% to 74% of the dry matter. Starch makes about 52-60% of the seed. It is present in the form of small granules about 1-2 μm in diameter (Chauhan et al., 1992). Other cereals and pseudocereals, such as rice, oat and amaranth, also contain small granule starches, with granules typically smaller than 5 μm in diameter. Several potential and existing uses for small granule starches have been described in the literature (Lindeboom et al., 2004). As the new food processing industries are increasingly, there is growing demand of both native and modified starches for the manufacture of various fabricated foods. This demand has created interest in finding the new sources of this starch. Starch has been used for moisture retention and to maintain the quality of stored food products. The limited publications on quinoa deal with its chemical composition, pasting properties and granule morphology a systematic documentation of functional, structural and textural properties of quinoa starch is still to be conducted. Hence, the aim of this study was to characterize the physicochemical, thermal, textural, and structural properties of quinoa starch.

II. MATERIALS AND METHODS

A. MATERIALS

The pure seeds of white quinoa (*Chenopodium Quinoa* Willd.) were procured from Devshree Grains & Pulses store, Khari Baoli market, Delhi. The quinoa seeds were cleaned and stored in refrigerator at 4°C.

B. STARCH ISOLATION

Quinoa seeds were steeped in sodium hydroxide (0.3%) at ambient temperature overnight followed by homogenization with a kitchen blender, the slurry was screened with a series of sieves, the last one with 0.125 mm of aperture size. The filtrate was centrifuged at 6000 rpm, then the supernatant was discarded, the small amounts of protein and fine fiber on the surface of the sediment were scraped off and cakes were reslurried in alkaline solution and centrifuged again, this operation was repeated for 3 times. Then deionized water and 95% aqueous ethanol were applied successively to rinse and desiccate starch, respectively. The final starch cakes were crushed and defatted with petroleum ether (b.p. 40-60 °C) by decanting the lipid containing supernatant for 3 times after steeping overnight. It was kept at ambient temperature for more than 2 days to reach moisture equilibrium.

C. PHYSICOCHEMICAL PROPERTIES OF STARCH

a. AMYLOSE CONTENT

The amylose content of isolated starch was determined by using the method Williams et. al. (1970). A starch sample (20 mg) was taken and 10 ml of 0.5 N KOH was added to it. The suspension was thoroughly mixed. The dispersed sample was transferred to a 100 ml volumetric flask and diluted to the mark with distilled water. An aliquot of the test starch solution (10 ml) was pipetted into a 50 ml volumetric flask and 5 ml of 0.1 N HCL was added followed by 0.5 ml of iodine reagent. The volume was diluted to 50 ml and the absorbance was measured at 625 nm. The measurement of the amylose content (%) was determined from a standard curve developed using amylose and amylopectin blends.

b. SWELLING POWER AND SOLUBILITY

Swelling power and solubility were determined over a temperature range of 65-95°C according to the method of Leach and McCowen(1959).

c. WATER SOLUBILITY INDEX (WSI) AND WATER ABSORBANCE INDEX (WAI)

WSI and WAI were determined using a modified method by Anderson (1981).

D. THERMAL PROPERTIES

The thermal characteristics of the starches were analyzed using Differential Scanning Calorimeter (PerkinElmer DSC 4000). Starch samples were weighed in an aluminum pan and distilled water was added with the help of a micro syringe to obtain a starch water suspension containing 70% water (w/w). The pan was hermetically sealed and allowed to equilibrate for 1 h. The instrument was calibrated using indium and empty aluminum pan was used as reference. Onset (T_o), peak (T_p) and conclusion (T_c) temperatures and enthalpy of gelatinization (ΔH_{gel} , J/g) were recorded.

E. TEXTURAL PROPERTIES

Textural properties of RVA starch gels were evaluated using the TA/XT2 Texture Analyzer. The starch prepared in the RVA were poured into small aluminum canisters and stored at 4 °C to cause gelation. The gel formed in the canisters was evaluated for their textural properties by texture profile analysis (TPA) using the TA/XT2 texture analyzer. Each canister was placed upright on the metal plate and the gel was compressed at a speed of 0.5 mm/s to a distance of 10 mm with a cylindrical plunger (diameter = 5 mm). The compression was repeated twice to generate a force-time curve from which hardness (height of first peak) and springiness (ratio between recovered height after the first compression and the original gel height) were determined. The negative area of the curve during retraction of the probe is termed adhesiveness. Cohesiveness was calculated as the ratio between the area under the second peak and the area under the

first peak (Bourne, 1968; Friedman, Whitney, & Szczesniak, 1968). Gumminess was determined by multiplying hardness and cohesiveness. Chewiness was derived from gumminess and springiness and was obtained by multiplying these two. Five repeat measurements were performed for each sample and their average was taken.

F. PASTING PROPERTIES

A Rapid Visco-Analyzar (RVA) was used to determine the pasting properties of starch samples according to AACC method 61-02 (AACC 2000). A 5% (w/v) starch slurry was made in the RVA canister which was kept at 50°C for 1 min then heated to 95°C in 3.8 min, held at 95°C for 1 min, and then cooled to 50°C within 3.8 min where it was held for 1.4 min. For the first 10 s of the test, the slurry was stirred at a speed of 960 rpm, and at 160 rpm for the remainder of the test.

G. STRUCTURAL PROPERTIES

a. SCANNING ELECTRON MICROSCOPY (SEM)

Scanning electron micrographs of starch granules were taken Scanning Electron Microscope (JEOL, JCM-6000). Starch samples were mounted on the aluminum studs using double sided adhesive tape and coated with gold. An accelerating potential of 10 kV was used during microscopy.

b. X-RAY DIFFRACTION ANALYSIS (XRD)

X-ray diffraction was performed using a MiniFlex Desktop X-ray diffractometer (Rigaku Co. Ltd.) operating at 40kV and 80 mA. Diffractograms were obtained from 10° 4θ to 70° 4θ with a scanning speed of 4°/min.

III. RESULTS AND DISCUSSION

A. PHYSIO-CHEMICAL PROPERTIES OF STARCH

a. AMYLOSE CONTENT

The fraction of amylose content in quinoa starch was 9.22% which is lower than the amylose contents of cereal, root, tuber and legume starches. Atwell et al.(1983) reported an amylose content of 11% for quinoa starch. The low amylose content and small starch granule size of quinoa starch significantly affects functional characteristics of the starch.

b. SWELLING POWER AND SOLUBILITY

Swelling power and solubility values for the quinoa starch are presented in Figure 1. Quinoa starch produced a much higher swelling power. As shown in the Figure 1(a), increments in the pasting temperature increased the swelling power of quinoa starch. The strength and character of micellar network within the granule is the major factor controlling the swelling behavior of starch. Thus, a highly associated starch with an extensive and strongly bonded micellar structure should be relatively resistant to swelling. Swelling power was

strongly affected by amylose content. It is accepted that amylose acts as a restraint to swelling (Tester and Morrison, 1990, Fredriksson et al., 1998; Sasaki et al., 2003; Noosuk et al., 2003) and waxy starches swell to a greater extent than their normal amylose counterparts (Tester and Morrison, 1990). Lorenz (1990) and Ahamed et al. (1996a) reported that quinoa starch had a high swelling power compared to wheat, barley, rice, amaranth, potato and corn starches.

Solubility of quinoa starch increased with higher pasting temperatures as shown in the Figure 1 (b). Solubility reflects the leakage of amylose from starch granules (Ahamed et al., 1996a). During pasting, it is mainly the amylose component which leaches out and quinoa starch was the lowest in amylose content as revealed in this study.

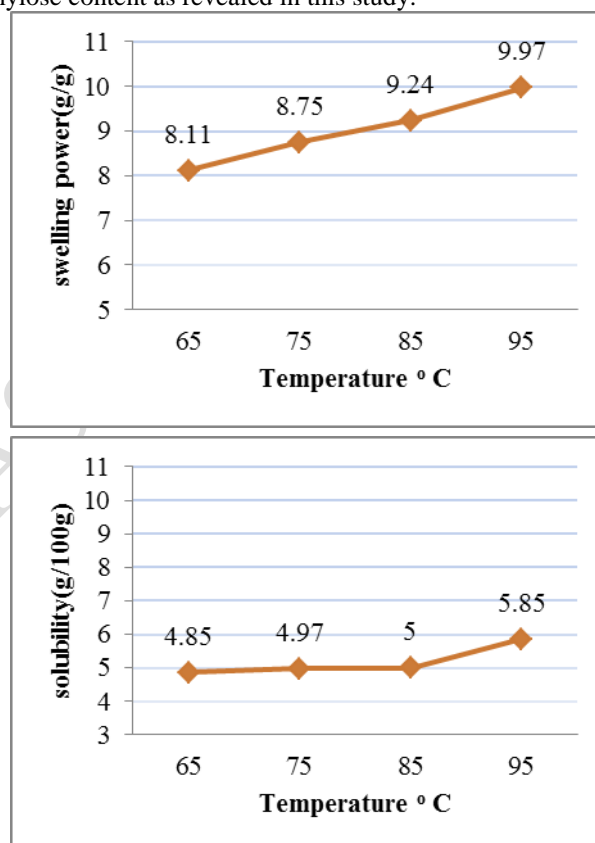


Figure 1: (a) Swelling power and (b) Solubility index of Quinoa starch

c. WATER SOLUBILITY INDEX (WSI) AND WATER ABSORBANCE INDEX (WAI)

Functional properties are the intrinsic physicochemical characteristics which may affect the behavior of food systems during processing and storage. Water solubility index and water absorbance index of quinoa starch were 1.13% and 2.23% respectively. The WAI of starch makes quinoa suitable to be used as a thickener in liquid and semi-liquids foods since the starch was able to absorb water and swell for improved consistency in food.

B. THERMAL PROPERTIES

The thermal properties of quinoa starch are presented in Table 1. Gelatinization temperatures studied were found to be

$T_0(50.36^\circ\text{C})$, $T_p(96.49^\circ\text{C})$, $T_c(146.00^\circ\text{C})$ and enthalpies of gelatinization (ΔH_{gel}) (275.36 J/g). The thermal characterization of starch is important for determining the cooking variables of quinoa grain starch. T_p gives a measure of crystalline quality (double helix length). The higher gelatinization temperatures of quinoa starch in study might be due to the lower amylose content of starch. According to Gernat et al. (1993) and Fredriksson et al. (1998), starch crystallinity increases with amylopectin content. Hence, starches with higher amylopectin contents would be expected to have higher onset, peak and conclusion temperatures. Enthalpy of gelatinization (ΔH_{gel}) is an indication of the loss of molecular order within the granule and it gives an overall measure of crystallinity (quantity and quality) (Cooke and Gidley, 1992; Hoover and Vasanthan, 1994; Tester and Morrison, 1990). In this study quinoa starch showed the high gelatinization enthalpy, suggesting that granule architecture has a high molecular order and longer amylopectin double helices.

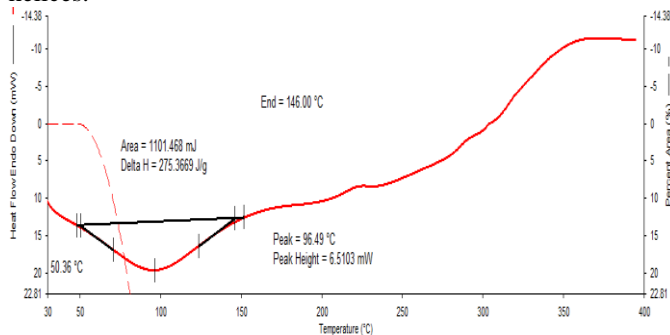


Figure 2: DSC profile of starch obtained from quinoa.

C. TEXTURE PROPERTIES OF STARCH GEL

The textural properties of gel from quinoa starch determined using texture analyzer are shown in Table 1. Starch gel from quinoa starch showed the hardness of 596.1 g/force. The starch gel firmness is mainly caused by retrogradation of starch gels, which is associated with the syneresis of water and crystallization of amylopectin, leading to harder gels (Miles et al., 1985). Mua & Jackson (1997) showed that starches that have harder gels tend to have higher amylose content and longer amylopectin chains. In this study the gumminess and chewiness was found to be 259.50 and 281.29, respectively. The mechanical properties of starch gels depend upon various factors, including the rheological characteristics of the amylose matrix, the rigidity and the volume fraction of the gelatinized starch granules, as well as the interactions between continuous and dispersed phases of the gel (Biliaderis, 1998). These factors are in turn depending on the amylose content and the structure of the amylopectin (Yamin et al., 1999).

Sample	Thermal Parameters				Gel Textural parameters					
	$T_0(^{\circ}\text{C})$	$T_p(^{\circ}\text{C})$	$T_c(^{\circ}\text{C})$	$\Delta H(\text{J/g})$	HD(g)	AD(g)	CH	RS	GM	SP
Quinoa	50.36	96.49	146.00	275.36	596.1	24.43	281.29	0.322	259.50	1.08

T_0 -onset; T_p -peak ; and T_c - conclusion temperatures and ΔH is heat of gelatinization.

HD- Hardness; AD- Adhesiveness; CH- Chewiness; RS- Resilience; GM- Gumminess; SP- Springiness.

Table 1: Thermal and Gel Textural properties of quinoa starch

D. PASTING PROPERTIES

Pasting profile of starch extracted from quinoa is presented in Figure 3. The pasting properties of starch are useful to obtain information of its functional behavior during heating and cooling periods of starch, which is common during the processing of starchy products or in those where starch is added as ingredient. Pasting temperature (PT) of starch extracted from quinoa was found to be 76.75°C . Pasting temperature is an indication of the minimum temperature required to cook the flour. The increase in viscosity with increase in temperature may be attributed to the removal of water from the exuded amylose by the granules as they swell (Ghiasi, Varriano-Marston, & Hosene, 1982). Final viscosity (FV) (indicates the ability of the starch to form a viscous paste) of quinoa starch was 4004.00 cP. Increase in final viscosity might be due to the aggregation of the amylose molecules (Miles et al., 1985). Setback viscosity (SB) (measure of syneresis of starch upon cooling of the cooked starch pastes or retrogradation tendency) of quinoa starch was 668.00 cP. The lowest setback viscosity of starch indicates its lower tendency to retrograde. Adebowale and Lawal (2003a) reported that the smaller tendency to retrograde are an advantage in food products such as sauces and soups, which undergo loss of viscosity and precipitation as a result of retrogradation. Breakdown viscosity (BD) (measure of the ease with which the swollen granules can be disintegrated) showed for quinoa starch was 257.00 cP.

Sample	PT($^{\circ}\text{C}$)	P _{time} (min.)	PV	TV	FV	BD	SB
Quinoa starch	76.75	5.33	3593.00	3336.00	4004.00	257.00	668.00

PT- pasting temperature; P_{time}- peak time; PV- peak viscosity; TV- trough viscosity; FV- final viscosity; BD- breakdown; SB- setback.

Table 2: Pasting properties of quinoa starch (viscosity in cP)

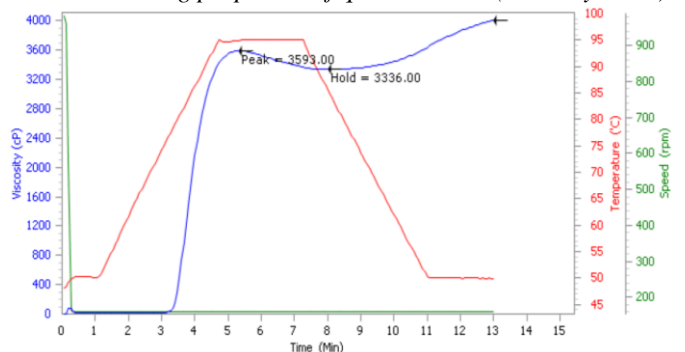


Figure 3: Rapid Visco Analyzer of quinoa starch

E. X-RAY DIFFRACTION ANALYSIS (XRD)

X-ray diffraction analysis have been useful in elucidating the structure of both amylose and whole starch. Native starch granules give distinctive X-ray diffraction patterns of three crystalline modifications designated A (cereal), B (tuber) and C (smooth pea and various beans) (Whistler et al., 1984).

Quinoa starch showed the typical A type X-ray diffraction pattern (Figure 4). The same patterns were reported by Paredes-Lopez et al. (1994) and Gorinstein and Lii (1992). The degree of relative crystallinity was 38.4 %. Jenkins and Donald (1995) reported that amylose disrupted the structure

order within the amylopectin crystallites. Thus, the crystallinity of starch is associated with the amylose content of starch.

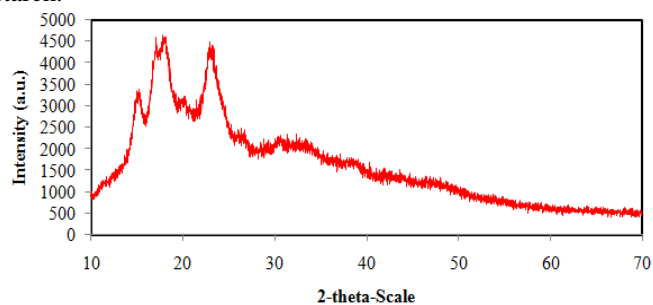


Figure 4: X-ray diffraction of quinoa starch

F. SCANNING ELECTRON MICROSCOPY

The Morphological characteristic of quinoa starch was determined by scanning electron microscopy. SEM of quinoa starch is presented in Figure 5. SEM observation showed that the quinoa starch particles were shaped irregular, polygonal and sized about 1-2 μm . In addition, granule surfaces of quinoa were less smooth than potato starch granules. Similar results were observed by Lindeboom et al. (2005), and Tang et al. (2002), in starches from different quinoa varieties. The small in size in comparison to corn starch, quinoa starch can act as carriers for flavors and colorants. (Zhao and Whistler, 1994).

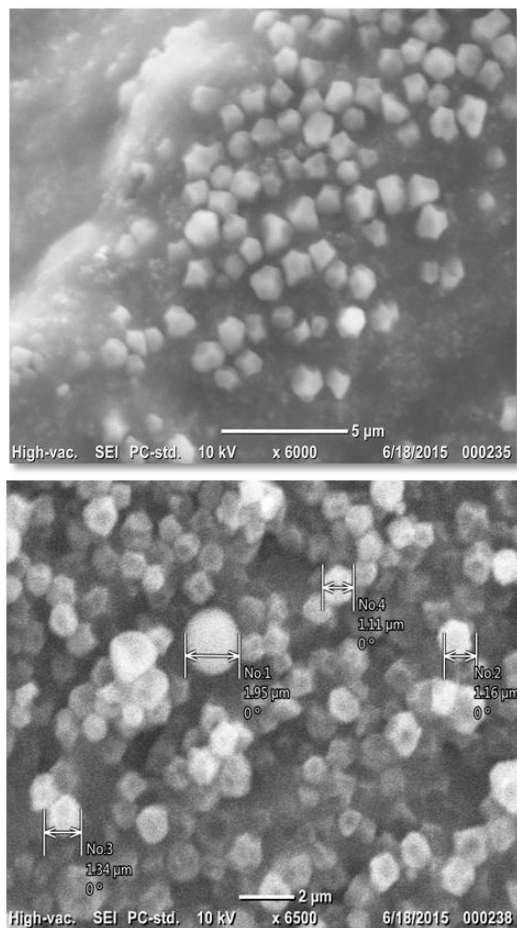


Figure 5: Scanning electron micrographs of quinoa starch

IV. CONCLUSION

The microscopy observation showed small granule trait of polygonal shape and size (about 2 μm) for quinoa starch. Quinoa starch had lower amylose content than other cereal starches. Quinoa starch exhibited a greater swelling power compared to wheat or barley starch. This study showed that as a thickening agent for fillings, quinoa starch performed better than other starches. The gelatinization temperature range of quinoa starch was higher than that of wheat and barley starch. It was found that gelatinization properties of quinoa starch differ significantly from other conventional sources of starch. Water solubility index and water absorbance index of quinoa starch was found 1.13% and 2.23% respectively. X-ray diffractometry revealed A-type patterns with higher crystallinity of quinoa starch. The pasting properties were studied by RVA (Rapid Visco Analyzer) which showed higher peak viscosity. In conclusion, quinoa starch properties indicated that this starch can be used for commercial starch production and used for various food (stabilizer, thickener and weaning foods) and non food applications such as the textile or pharmaceutical industries. Quinoa starch is suited for applications requiring improved binding and reduced breakability because of small granule diameter. The swelling power of quinoa starch places it in category of highly restricted swelling starch which is desirable for products like composite blends and noodles. Thus, quinoa starch could be used as a novel food source.

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