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Isolation and physico-chemical and rheological characterisation of the Brazilian jalap starch (Operculina tuberosa Meisn.)

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ABSTRACT

The properties of the jalap starch (Operculina tuberosa Meisn.) were investigated and compared with other already known starches (potato and wheat starch). The jalap starch presented peak viscosity lower than the one from potato but higher than wheat starch, while the stability during the cooling down was higher than potato and wheat starch. The jalap starch presented X-ray pattern of type-A, which is typical of those from wheat starch. The rheological and physico-chemical characteristics presented by this source of starch were intermediate between those from wheat and potato, which makes it a promising commercial source to be explored, mainly in areas with food scantiness as in the Brazilian Northeast.

1. Introduction

The jalap (Operculina tuberosa Meisn.) is a "climber" which belongs to the *Convolvulaceae* family and is common in the secondary vegetation of coastal areas (Matos, 1998). They are found in tropical regions comprised between Antilles and Brazil and temperate regions of the Mexicans Andes, in muddy areas and areas of deep soil (Planchon & Bretin, 1937). This family of 55 genuses include 1650 species of herbs, shrubs, many of which are aerial (Enriquez et al., 1992; Noda, Miyahara, Kawasaki, & Okabe, 1987; Shellard, 1961). One of the most interesting characteristics of this family is the presence of arrays of secretory cells of glycosidic resins in foliate tissue and, especially, in its roots. These substances constitute one of the chemotaxonomics characteristic of this family, and the employment in the traditional medicine of some species (*Convolvulus, Exogonium, Ipomoea, Merremia* and *Operculina*) is associated to the purgative properties of its resins (Garcia-Argáez & Pérez-Amador, 1997; Pereda-Miranda & Bah, 2003; Pérez-Amador, García-Argáez, Contreras, Herrera, & Ríos, 1998). The jalap resin is composed of two distinct types: jalapin and *jalapurgin* or *convolvulin*. The *jalapurgin*, the main active component, is an odorless, white glycoside which is more irritable than the *jalapin* and in large doses can act as poison (Culbreth, 1927).

The Brazilian jalap has as its main species *Operculina macrocarpa* Urb. (syns *C. macrocarpus* L., *O. macrocarpa* (L.) Farwel), *O. alata* (Ham) Urban whose synonym is *I. operculata* (Matos, 1997, 1998; Ono, Kubo, Miyahara, & Kawasaki, 1989), and I. tuberosa L. or *O. tuberosa* Meissner (Matos, 1997, 1998). All those species are known popularly as "*batata de purga*" (purge potato). It is used due to its laxative and purgative properties, against skin diseases and in the treatment of the leukorrhea (Martins, Castro, Castelani, & Dias, 2000; Matos, 1982; Michelin & Salgado, 2004). It is also used as menstruation regulator (Michelin & Salgado, 2004).

The roots of O. *tuberosa* can reach up to 40 cm in length, as was the case in this study. They are hard and difficult to break or crunch, and are found mainly in the Brazilian Northeast, where the roots are big, starchy and rich in resins (Matos, 1998).

The jalap powder sold commercially is rich in starch. However, its starch is considered of less interest by virtue of the importance of its resins. Phytochemical investigations on the resin glycosides were initiated during the second half of the XIX century. However, the structures of their active components had remained poorly known and still are for some members of these purgative root species (Pereda-Miranda & Bah, 2003). Scientific researches about the physico-chemical properties of jalap starch are unknown in the literature. This study has the objective to investigate the physico-

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chemical and rheological properties of the jalap starch, comparing it with other known sources of starch.

2. Materials and methods

2.1. Raw material

The jalap roots (O. tuberosa Meisn.) were collected in the rural zones of São Luís/MA (Brazilian Northeast) during the months of May and June. For the purpose of comparison, commercial wheat (*Triticum aestivum*) and potato (*Solanum tuberosum* L.) starches were used.

2.2. Isolation and purification of the starches

The roots were washed, peeled and grated manually. Enough water was added to allow the sieving of the mass rich in starch in sieves of 200 "mesh". After this stage the suspension was left to decant for 24 h and the red coloured supernatant was discarded and the sediment washed several times over until the white powder was obtained. The sample was dried in a stove with forced air circulation at 40 °C until humidity of approximately 10% was reached.

2.3. Chemical composition of starches

The jalap starch was submitted to moisture and ash analysis according to the methods 930.15 and 942.05 of AOAC (1990), respectively. The total nitrogen content was determined in micro Kjeldahl equipment, and the protein content was calculated by the conversion factor 5.75, in agreement with method 984.13 of AOAC (1990). The total lipids content was determined gravimetrically in agreement with method 954.02 of AOAC (1990) with small modifications after extraction with methanol in Soxhlet apparatus. The amylose content was determined according to the method 6647 of ISO (1987).

The phosphorus content (P) was determined as described in Zakharov, Motyguilin, and Girmutdinov (2000), with some modifications. Experiments were performed on an atomic absorption spectrometer 1100B (Perkin-Elmer,) with a deuterium background corrector, an HGA-700 atomizer, and an AS-70 autosampler. The light source was a phosphorus hollow-cathode lamp. Atomic absorption was measured at the wavelength of 213.5 nm, and the spectral slit width was 0.7 nm. Graphite furnaces with a pyrolytic cover and pyrographite platforms from Perkin-Elmer were used throughout.

2.4. Swelling power and solubility

Swelling power and solubility of starch sample were determined by the procedure described by Schoch (1964). About 1 g of starch and 40 mL of water were submitted to heating for 30 min at 55, 65, 75, 85 and 95 °C. After cooling, the slurry of starch was centrifuged 5000g for 30 min. The supernatant was carefully separated from the precipitate, and 10 mL was removed to evaluate the percent of soluble, while the sediment was weighed for determination of the percentage of swollen granules.

2.5. Pasting characteristics

A Rapid Visco Analyser (RVA) RVA-4 (Newport Scientific Pty. Ltd., Australia) was used for pasting determinations. About 4.5 g of starch were mixed with 25 mL of distilled water to obtain a total weight of 29.5 g in the RVA cup. The samples were maintained for 1 min at 50 °C and the temperature increased progressively up to 95 °C (13 °C/min.), held at 95 °C for 3 min. then cooled to 50 °C (13 °C/min.) and held for 3 min. The test was concluded after 13 min. The analyses were run in triplicate.

2.6. Gelatinization

The T_o (onset temperature), T_p (peak temperature), T_c (final temperature) and ΔH (gelatinization enthalpy) parameters, were obtained by analysis in a differential scanning calorimeter DSC-50 (Shimadzu, Japan). Approximately 3 mg of starch were weighed in aluminium capsules and suspended in 15 μL of distilled water to obtain the starch/water proportion of 1:5 (w/w). The samples were then placed to rest for 1 h and the sealed capsules heated from 25 to 100 °C (10 °C/min.).

2.7. Granule morphology

2.7.1. Optic microscopy

The granule morphology was studied by a polarized and bright-field optics microscope, (Jenalab Pol, Carl Zeiss JENA, Germany), from the Microscopy Laboratory of the Biology Department of Federal University of Ceará. The starch powder was suspended in aqueous solution of glycerol and a drop was spread in a microscope slide and re-covered with a cover slip. The samples were observed at a magnification of 50× in polarized and bright-field optics and photographed with a digital camera.

2.7.2. Scanning Electron Microscopy (SEM)

The morphology and the size of the granules were determined with the use of a scanning electron microscope Philips XL-30 (Philips, Eindhoven, Netherlands), from the Microscopy Laboratory of the Mechanical Engineering Department of UFC. The samples were mounted on aluminium stubs and coated with a 50 μm thick gold film in an Emitech K550 Sputter Coater, operating at 5 mA and 50 kV. Starch granule diameter range was estimated by measuring 10–20 randomly selected granules from triplicates microphotographs.

2.8. X-ray diffraction

The analysis was carried out in an X-ray diffractometer Dmax-B (Rigaku, Japan), with a copper radiation in line Kα ($\lambda = 0.1542$ nm), operating at 40 KV and 25 mA. The region of scanning of the diffraction angle (2θ) was 3–40° (1/2°/min.).

3. Results and discussion

3.1. Isolation and chemical composition of the starches

The jalap is a hard and fibrous root. This characteristic and harvest time affects enormously the starch extraction yield, which was low, around 12% (g starch/g fresh roots). Another parameter which probably also influenced the starch extraction yield was the high amount of resins impregnated in starch powder, which made the purification process, difficult.

The results of chemical composition of the starches are shown in Table 1. The amylose content was found to be inside the normally verified range for the starch of roots, which was near to those of the cassava, but lower than that of the wheat.

The phosphorus content in the jalap starch was low in relation to wheat and potato starches. When compared to cereals, legume and tubers starches and potato starch, were the only ones to have relatively high phosphorus content. The majority of those are in phosphate ester form substituted in anhydroglucose molecules of amylopectin. The presence of phosphate groups in amylopectin

Table 1
Chemical composition of the Jalap, wheat and potato starches.[a]

Determination	Wheat	Potato	Jalap
Moisture (%)	11.4 ± 0.4	12.1 ± 0.1	11.3 ± 0.3
Ashes (%)[b]	0.2 ± 0.0	0.4 ± 0.0	0.1 ± 0.0
Proteins (%)[b,c]	0.6 ± 0.0	0.2 ± 0.0	0.1 ± 0.0
Lipids (%)[b]	1.2 ± 0.3	0.1 ± 0.0	0.5 ± 0.0
Amylose (%)[b]	28.6 ± 0.2	27.0 ± 0.1	21.2 ± 0.0
Phosphorus (ppm)	51.0 ± 2.0[d]	110.0 ± 0.6	46.5 ± 0.6

[a] Mean of three determinations.
[b] Determinated in dry basis.
[c] $\%N \times 5.75$.
[d] Tester and Sommerville (2000).

influenced the thermal and rheological properties of these starches (Haase & Plate, 1996).

3.2. Microscopy of polarized and simple light

Polarization microscopy revealed the crystalline organization of the granules, as shown by the strong birefringence in Fig. 1. The granules presented the maltese cross very defined, indicating the presence of an intact hilum.

3.3. Scanning Electron Microscopy

The jalap starch granules, observed by SEM (Fig. 2a), demonstrates essentially polygonal, spherical and semi-spherical truncated forms, which vary in size in the ranges: 9.2–19.5 μm, 11.5–3.0 μm and 6.0–15.5 μm, respectively. These forms are probably results of the dissociation of the compound granules during the starch extraction. The truncated area is characterised by flat, concave or convex surfaces (Fig. 2b). The majority of the granule surfaces appeared smooth, without pores or fissures.

3.4. Swelling power and solubility

Fig. 3 shows that the root starches swell at lower temperatures and to a higher degree than those of the cereal starches. This shows that the degree of association of the root starches is smaller than in the cereal starches. The fast and intense swelling on the potato starch is attributed, in part, to the well pronounced presence of the phosphate groups in the amylopectin molecule, which increases the hydration, due to the electrostatic repulsion and absence of lipids (Galliard & Bowler, 1987). On the other hand, the wheat and the jalap, which possess greater lipid content and lower phosphate content, showed smaller swelling. The swelling of jalap starch increased when the temperature increased, more extensively than the wheat starch. This characteristic is because at 95 °C the granules reached its maximum swelling (absorption of water) until the total disruption of the granular structure. This suggests that the bonding forces of the jalap molecules are stronger than in the potato and smaller than in the wheat starch. The amylose and amylopectin content play an important role in the swelling of starches. The gelatinization and swelling properties are controlled in part by the molecular structure of amylopectin, starch composition and granule architecture of starches (Morrison, Tester, Snape, Law, & Gidley, 1993; Tester, 1997; Tester & Morrison, 1990); while the formation of amylose-lipids complex inhibit the swelling of the granule.

The solubility of the jalap was intermediate when compared to potato and wheat starch. Up to 65 °C the values were almost the same. However, from 75 °C, the jalap obtained higher solubility in relation to the wheat. This behavior can be due to higher content of lipids complexed with amylose in wheat starch which decreases the leaching out of amylose from the granules and consequently the solubility of the starch.

At 85 °C it was not possible to obtain the solubility and swelling degree of potato starch because it was not observed a clear separation between the solubilised and swelled material, which provided a single phase (Fig. 3).

3.5. Pasting properties

Table 2 demonstrates that the tubercles and roots have an elevated peak viscosity (PV) more conspicuous than the wheat (a cereal starch). The PV of jalap starch is high when compared to that of the wheat starch, but much lower than the one from potato. These differences can have various causes, the main being the amylose content and the presence of phosphate monoesters groups. It is well established in the literature that roots and tuber starches contain significant amount of phosphate monoesters covalently bound to starch (Kasemsuwan & Jane, 1995).

The high PV of potato starch is caused in part by the polyelectrolyte effect of the phosphate monoester groups on the amylopectin molecules and by the absence of lipids (Greenwood, 1976). On the other hand, the swelling of the wheat starch is suppressed by the presence of lipids complexed with amylose (Banks & Greenwood, 1975). The PV of jalap starch is intermediate between the two. It is difficult to explain this behaviour as the nature of P present in this starch is not known yet.

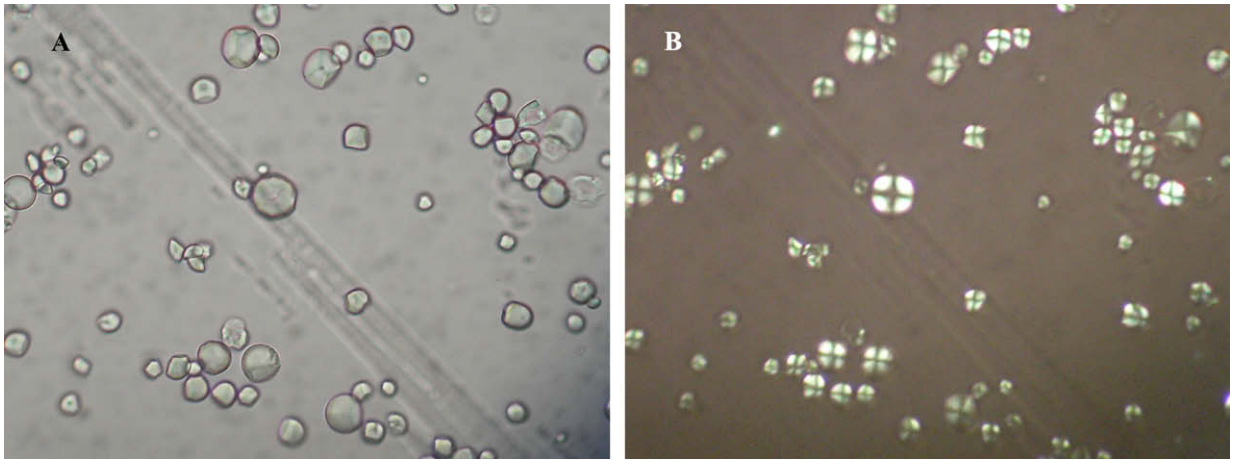

Fig. 1. Optic microscopy of jalap starch granules viewed with bright-field (A) and polarization optics (B).

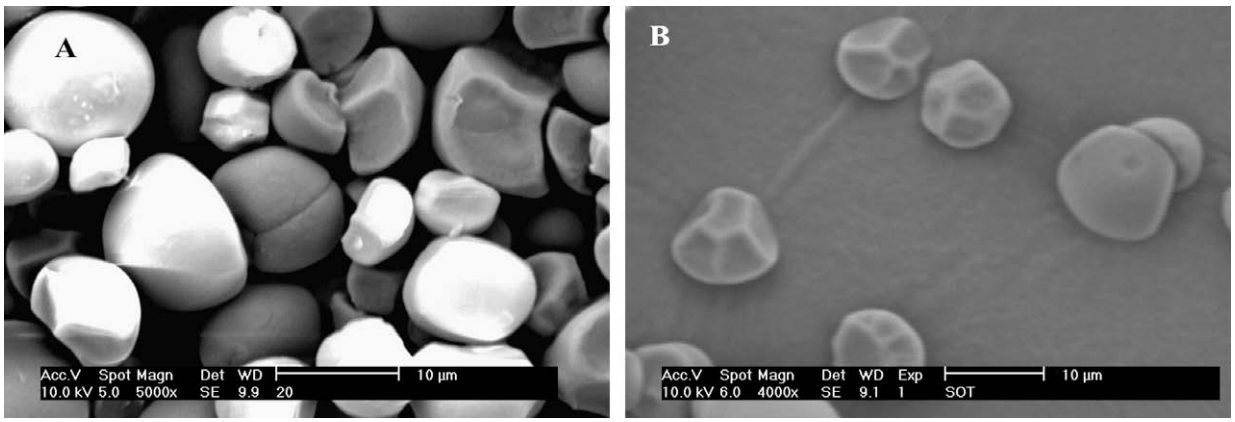

Fig. 2. SEM of the jalap starch. (A) Overview of granules and (B) individual granule aspects.

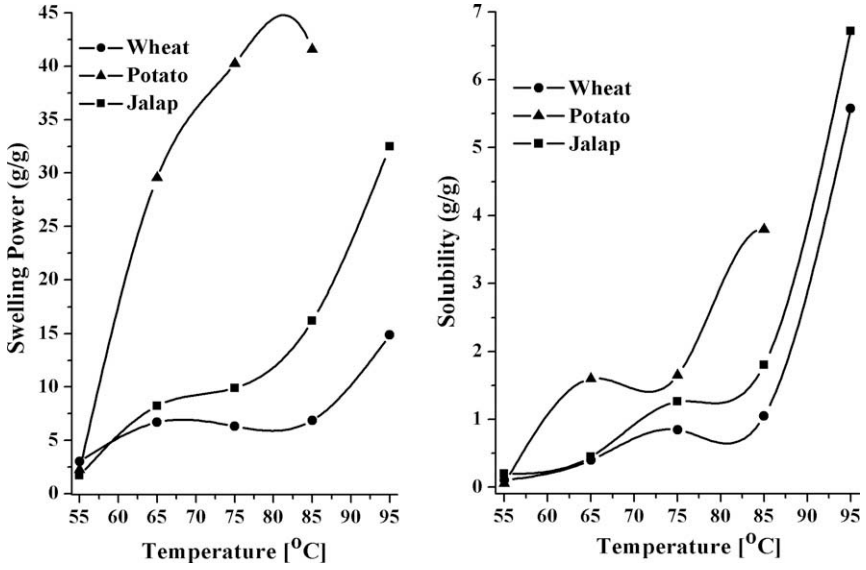

Fig. 3. Swelling and solubility curves of jalap, potato and wheat starches.

Table 2
Pasting characteristics of native wheat, potato and jalap starches.[a]

Sample	Peak Visc. (cP[b])	Breakdown[c] (cP)	Hold (cP)	Setback[c] (cP)	Final Visc. (cP)	Pasting Temp. (°C)
Wheat	2872	760	2113	1096	3210	86.4
Potato	10173	7160	3014	483	3497	67.8
Jalap	4328	1312	3017	863	3880	76.0

[a] Mean of three determinations.
[b] cP, centPoise.
[c] Breakdown, peak visc.-hold; setback, final visc.-hold.

The viscosity of the hot paste (hold) and the final viscosity of the jalap starch were similar to the potato. On the other hand, the tendency to retrograde, as deduced from the setback, was almost doubled. This may be a consequence of the size and organization of the molecules of starch. However, we cannot affirm this with certainty.

Fig. 4 demonstrates that when the maximum temperature of gelatinization is reached (95 °C), the granules suffer a high degree of swelling, resulting in a high peak viscosity. This behaviour can be observed in the swelling power analysis at 95 °C (Fig. 3). There is a similarity between RVA and swelling power analysis at 95 °C, which is the temperature in which the total gelatinization of the granules begins. After this stage, the gel formation takes place.

The increase in viscosity when the hot starch paste is cooled depends on the retrogradation tendency of the starch, which is related to the amount and molar mass of amylose fraction (French, 1975; Jane et al., 1999). The retrogradation rate decreases with longer and shorter amylose molecules, having the optimum length as reference. Long amylose molecules do not readily move into tight association with other chains and have difficulty lining up with their neighbors over long intervals. Amylose molecules smaller than the optimum length do not associate completely and are too short to form a gel (Fennema, Powrie, & Marth, 1973; Lawrence, 1985; Swinkels, 1985). Suzuki, Takeda, and Hizukuri (1985) studied the relation between the retrogradation rate and the molecular properties of the starch and suggested that the high-

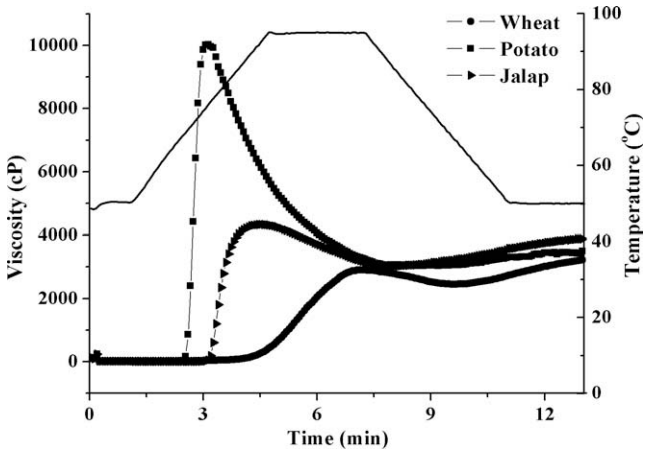

Fig. 4. RVA curves of jalap, potato and wheat starches.

er the molecular average weight the smaller the tendency to retrogradate.

The pasting characteristics of the jalap starch showed a high viscosity of hot paste and larger paste stability on cooling and low tendency to retrogradation. These characteristics are extremely desirable in applications such as thickening in dehydrated soups and sauces, binding in sausages, stabilizer in salad sauces and gelling in puddings and desserts (Cereda, 2002; Silva et al., 2006).

3.6. Gelatinization temperature

The gelatinization peak of the wheat starch (128.0 °C) occurred at a higher temperature than that potato (108.6 °C) and jalap (114.1 °C) starches (Fig. 5), probably an influence of lipids and protein contents (Table 1). The gelatinization enthalpy jalap starch (ΔH = 10.2 J/g) was higher to those of wheat (8.1 J/g) and smaller than potato (10.8 J/g).

Noda, Takahata, Sato, Ikoma, and Mochida (1996) have postulated that DSC parameters are influenced by the molecular architecture of the crystalline region, which corresponds to the distribution of amylopectin short chains (DP 6-11) and not by the proportion of crystalline region which corresponds to the amylose to amylopectin ratio. They showed that in sweet potato and wheat starches the low gelatinization parameters (T_o, T_p, T_c and ΔH) reflected the presence of short amylopectin chains.

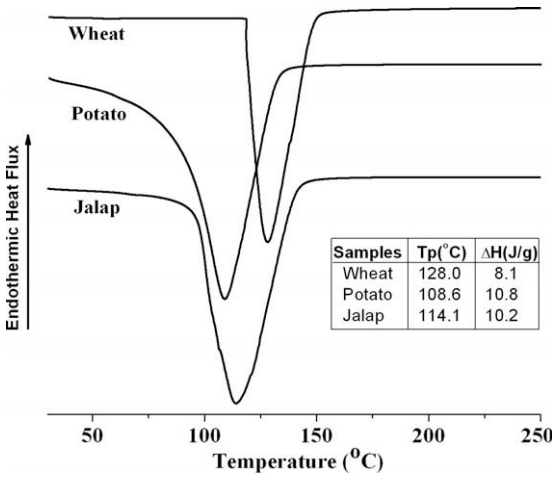

Fig. 5. DSC curves of jalap, potato and wheat starches.

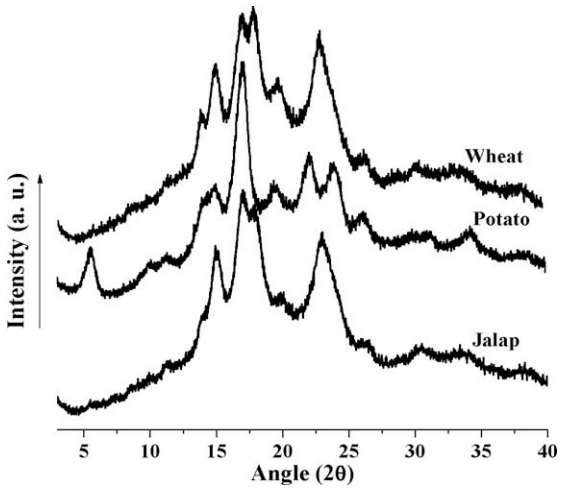

Fig. 6. X-ray diffraction patterns of native jalap, potato and wheat starches.

Jenkins et al. (1994) suggested that gelatinization in excess of water is primarily a swelling driven process. The swelling process destabilize the amylopectin crystallites within the crystalline lamellae, which are ripped apart (smaller crystallites are destroyed first). This process occurs rapidly for an individual crystallite, but takes much longer for the whole granule. The T_p and ΔH values for the jalap starch suggest that the size of the crystal inside the granules is of higher order than those from wheat and very similar to potato starches.

3.7. X-ray diffraction

The X-ray patterns of the potato starches, wheat and jalap are shown on Fig. 6. The potato starch demonstrates a pattern type-B, with the characteristic peak of this in 5^o (2θ). However wheat and jalap starches presented a pattern type-A which is characteristic of cereal starches. This X-ray pattern is characterised by a double helical packing, which have fewer water molecules in its unit cell than B-type starch like potato starch.

4. Conclusions

The jalap starch showed some physico-chemical characteristics intermediate between wheat and potato starches. This can lead us to assume a promising future from this source of starch in the food industry as thicker, stabilizer and gelling and as carbohydrate source for human nutrition especially in areas not favorable to the cultivation, as this root is not very demanding as far as soil and humidity are concerns.

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