Available online at www.sciencedirect.com
SCIENCE DIRECT
Carbohydrate Polymers 61 (2005) 111–114
Carbohydrate Polymers
www.elsevier.com/locate/carbpol

Short communication

New starches from *Fritillaria* species medicinal plants

Shujun Wang[a], Wenyuan Gao[a,*], Haixia Chen[a], Peigen Xiao[b]

[a]College of Pharmaceuticals and Biotechnology, Tianjin University, Tianjin 300072, China
[b]Institute of Medicinal Plant, Chinese Academy of Medical Sciences and Peking Union Medical College, Beijing 100094, China

Received 6 December 2004; revised 2 March 2005; accepted 30 March 2005
Available online 17 May 2005

Abstract

Fritillaria, an important medicinal plant, are widely utilized in the pharmaceutical industry. The bulbs of *Fritillaria* contain plenty of starches which have been ignored and wasted during the isolation and separation of the small-molecule bioactive ingredients. With the development of food and medicine industry, new starches with different properties have been paid more and more attention. The object of this work is to study the basic physicochemical properties of starch contained in the *Fritillaria* bulbs. The results revealed that the crystal type of the three *Fritillaria* starches was the characteristic B-type which was in agreement with the crystal type of potato starch. The results of scanning electron microscope showed that the size of the *Fritillaria thunbergii* and *Fritillaria ussurensis* starch granules ranged from 5 to 30 μm, while the *Fritillaria pallidifloca* starch granules ranged from 5 to 40 μm. The shape of the three *Fritillaria* starches was also different from each other. The starch separated from *F. ussurensis* showed the highest enthalpies of gelatinization (ΔH_{gel}) and the lowest transition temperature (T_o, T_p, and T_c), while the starch from *F. thunbergii* showed the highest transition temperature, peak height indices (PHI) and the lowest gelatinization temperature range (R).

Keywords: *Fritillaria*; Starch; Crystallography; Morphology; Thermal

1. Introduction

Starches, the most important polysaccharide reserve in higher plants, from different food plants sources, such as corn, rice, wheat, and potato, have been studied for several centuries. However, the starches contained in the medicinal plants have hardly been studied and are currently wasted during the isolation and separation of the small-molecule ingredients. With the developments in the food and pharmaceutical industries, new starches with distinct properties have been paid more and more attention.

The bulbs of *Fritillaria* species, Beimu, have been used as one of the most important anti-tussive, expectorant, and anti-hypertensive drugs in traditional Chinese medicine (TCM) for thousands of years. Various chemical and pharmacological studies on Beimu have demonstrated that the major biologically active ingredients to relieve cough in the bulb are alkaloids with their types and contents varying in different *Fritillaria* species (Li et al., 1999; Li, Li, Lin, Chan, & Ho, 2000; Li, Li, & Lin, 1999; Li, Lin, Chan, & Li, 2001; Lin, Li, Li, & Chan, 2001). In addition, some non-alkaloid constituents containing saponin, terpenoids, steroids, succinic acid, thymidine, adenosine in different *Fritillaria* species have also been identified (Ruan, Zhang, & Wu, 2002), however, the starch contained in the bulb of *Fritillaria* has hardly been studied and all wasted. As we reported earlier, the main component in the bulbs of *Fritillaria* species is starch occupying approximately 80% content in the total biomass (Gao, Fan, & Paek, 1999).

In addition to potential industrial application of starch from medicinal plants here, study is of interest as it provides some evidence for the medicinal plant chemical taxonomy from a macromolecular aspect. Chemical taxonomy of medicinal plant is primarily based on the small-molecule chemical constituents (Yu & Xiao, 1992), however, starch, a primary metabolite of the plant, is more easily to reflect the taxonomic characteristics of medicinal plant.

The objective of this work is to study the three *Fritillaria* starches for food and medicine industry. In addition, it also could provide some macromolecule information on the plant chemical taxonomy in the future.

* Corresponding author. Tel.: +86 22 87401895.
E-mail address: edwinwa@hotmail.com (W. Gao).

0144-8617/$ - see front matter © 2005 Elsevier Ltd. All rights reserved.
doi:10.1016/j.carbpol.2005.03.017

2. Materials and methods

Fritillaria thunbergii Miq., *Fritillaria ussurensis* Maxim. and *Fritillaria pallidifloca* Schrenk were provided by Meiwei TCM company (Anguo, Hebei province, China) and were identified by Professor Xiao Peigen, Institute of Medicinal Plant, Chinese Academy of Medical Sciences and Peking Union Medical College, China.

In the process of starch extraction, the three *Fritillaria* were cleaned, comminuted to powders and passed through a 160 mesh sieve. The *Fritillaria* powders were extracted following by the constant stirring with 85% alcohol at 45 °C for 48 h. The residue was washed with 85% alcohol several times until the supernatant liquid was transparent, and then desiccated at ambient temperature for further use.

For SEM analysis, starch samples were suspended in acetone to obtain a 1% suspension. One drop of the starch–acetone suspension was applied on a glass slide. After the acetone volatilized, the samples were then coated with gold powder to avoid charging under the electron beam and analyzed for starch granule shape and size using an environmental scanning electron microscope (ESEM, Philips XL-3). An accelerating potential of 30 kV was used during micrography.

Crystallography of the three *Fritillaria* starches was studied by a BDX3300 X-ray powder diffractometer. The starch powders were packed tightly in a rectangular aluminum cell. The samples were exposed to the X-ray beam from an X-ray generator running at 36 kV and 20 mA. The scanning regions of the diffraction angle 2θ were 10–30°, which covered most of the significant diffraction peaks of the starch crystallites. Other operation conditions included: step interval 0.02, scan rate 2°/min, Sollet and divergence slit 1°, receiving slit 1°, and scattering slit 0.15°. Duplicate measurements were made at ambient temperature. Radiation was detected with a proportional detector. The degree of crystallinity of three *Fritillaria* starches was quantitatively estimated following the method of Nara and Komiya (1983) (Cheetham & Tao, 1998).

Thermal characteristics of isolated starches were studied by using a differential scanning calorimeter-DSC204, HP (NETZSCH, Germany) equipped with a thermal analysis station. Starch (3.5 mg, dry weight) was loaded into a 40 μl capacity aluminium pan (Mettler, ME-27331) and distilled water was added with the help of Hamilton microsyringe to achieve a starch–water suspension containing 70% water. Samples were hermetically sealed and allowed to stand for 1 h at room temperature before heating in the DSC. The DSC analyzer was calibrated using indium and an empty aluminium pan was used as reference. Sample pans were heated at a rate of 10 °C/min from 20 to 120 °C. Onset temperature (T_o), peak temperature (T_p), conclusion temperature (T_c), and enthalpy of gelatinization (ΔH_{gel}) were calculated automatically. The gelatinization temperature range (R) was computed as ($T_c - T_o$) as described by Vasanthan and Bhatty (1996). Enthalpies were calculated on a starch dry basis. The peak height index (PHI) was calculated by the ratio $\Delta H/(T_p - T_o)$ as described by Krueger, Knutson, Inglett, and Walker (1987).

3. Results and discussion

SEM showed that the *F. thunbergii* and *F. ussurensis* starch granules were predominantly oval and elliptic, with visible thickness and varying granule size (Figs. 1 and 2), and with no obvious differences between the two species. The granule size of the two *Fritillaria* starches ranged from 5 to 30 μm. The average particle size ranged from 5 to 15 μm for small and 20–30 μm for large granules. These two *Fritillaria* starches showed the presence of a fairly large number of large-sized, elliptic-shaped granules. However, the granule size of the *F. pallidifloca* starch was much variable and ranges from 5 to 40 μm. The average granule size of starch ranged between 5 and 20 μm for small and 25–40 μm for large granules. The shape of starch granule was oval, elliptic, and polygonal. The surface of the starch granule was some unsmoothed and uneven. Much different from the above two *Fritillaria* starches, the *F. pallidifloca* starch had the small-sized granule in a much larger number. Granule size and shape are a characteristic of the plant starch. The microscopic structure of the plant starch granules may influence their industrial application.

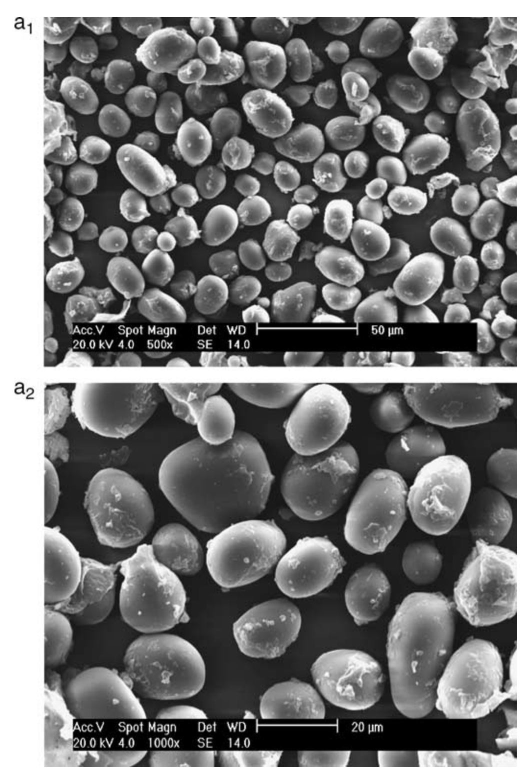

Fig. 1. SEM of *F. thunbergii* starch granules.

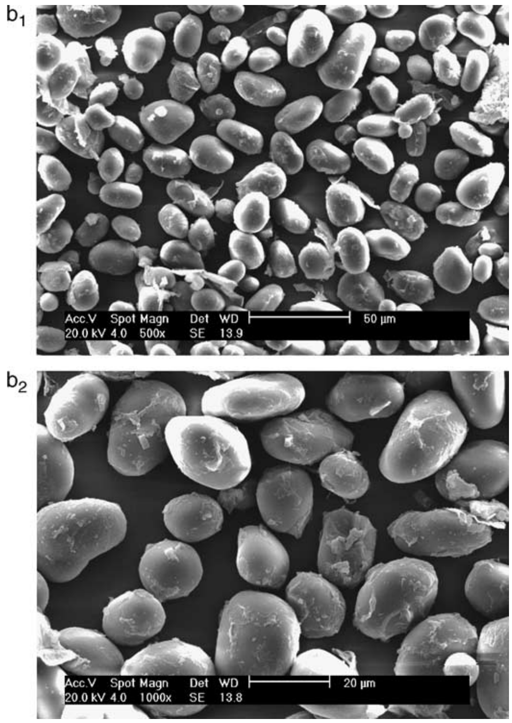

Fig. 2. SEM of *F. ussurensis* starch granules.

The X-ray diffraction patterns were compared with the reported standard diffraction patterns of different crystalline types (Zobel, 1964, 1988). The X-ray diffraction patterns of the three *Fritillaria* starches were presented in Fig. 3.

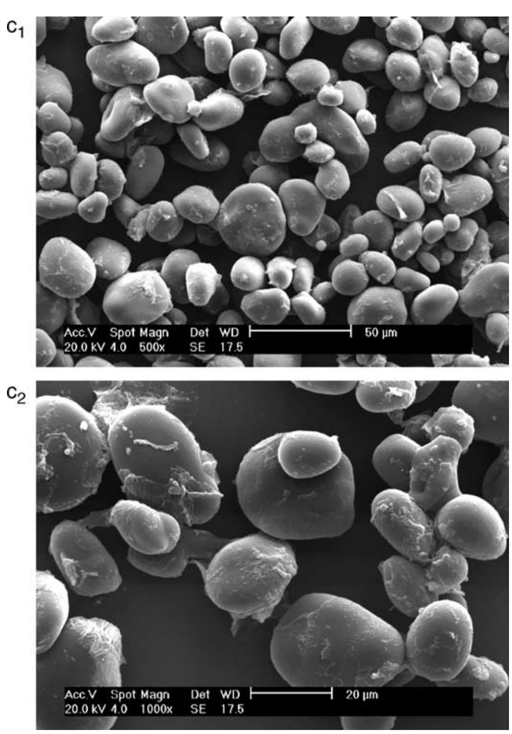

Fig. 3. SEM of *F. pallidifloca* starch granules.

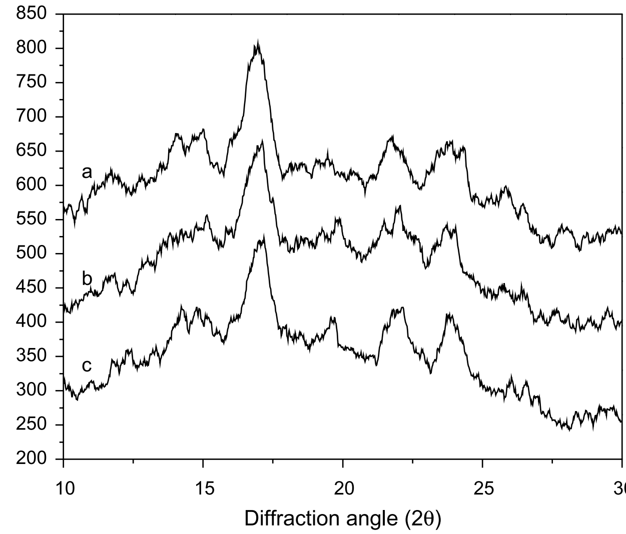

Fig. 4. X-ray diffraction patterns of the three *Fritillaria* starches.

As was well known, potato starch showed the characteristic B-type pattern with strongest reflection at 2θ about 17° and a few small reflections at around 2θ values of 15, 20, 22, and 24°. The three *Fritillaria* starches also gave the strongest diffraction peak at around 17° 2θ and a few small peaks at around 2θ values of 15, 20, 22, and 24°. This result demonstrated that the crystal type of the three *Fritillaria* starches was also a characteristic B-type.

The degree of crystallinity of the three *Fritillaria* starches calculated from Fig. 4 was shown in Table 1. For this evaluation, starches with the same moisture contents (∼16%) were used in order to minimize the effect of different moisture contents on crystallinity.

The transition temperatures (T_o, T_p, and T_c), range ($T_c - T_o$), enthalpies of gelatinization (ΔH_{gel}), and peak height indices (PHI) of starches from different *Fritillaria* differ significantly (Table 2). *F. ussurensis* starch showed the highest ΔH_{gel} value of 9.12 J/g and *F. pallidifloca* starch showed lowest ΔH_{gel} value of 6.87 J/g. *F. thunbergii* starch had the highest T_o (67 °C), followed by *F. pallidifloca* starch (63.9 °C), while it was lowest for *F. ussurensis* starch (62.1 °C). *F. thunbergii* starch showed the highest T_p and T_c of 71.7 and 78 °C, respectively. *F. ussurensis* starch showed the lowest T_p and T_c of 67.2 and 75.5 °C. *F. thunbergii* starch showed the maximum PHI and the minimum R values. These different thermal properties of starches from different *Fritillaria* may be attributed to the differences in granule structure, amylose content, the gelatinization

Table 1
X-ray diffraction data of the three *Fritillaria* starches

Samples	Degree of crystallinity (%)	Crystal pattern
F. thunbergii	37.8	B
F. u*ssurensis*	33.9	B
F. pallidifloca	41.8	B

Table 2
Thermal properties of starches separated from different *Fritillaria*

Samples	T_o (°C)	T_p (°C)	T_c (°C)	ΔH_{gel} (J/g)	PHI	R
F. thunbergii	67	71.7	78	8.82	1.88	11.0
F. ussurensis	62.1	67.2	75.2	9.12	1.79	13.1
F. pallidifloca	63.9	70.5	76.1	6.87	1.04	12.2

T_o, onset temperature; T_p, peak temperature; R, gelatinization range ($T_c - T_o$); ΔH_{gel}, enthalpy of gelatinization (dwb, based on starch weight); PHI, peak height index $\Delta H_{gel}/(T_p - T_o)$.

temperature, and the presence of crystalline regions of different strength in the granules (Singh & Singh, 2001).

4. Conclusions

Starches from medicinal plant are now first studied by commonly used methods. *Fritillaria* having high starch content may be considered as new starch sources for the food and medicine industry. Due to the differences in the thermal properties compared with other starches, especially with low ΔH_{gel}, PHI, and high R value, *Fritillaria* starch could provide more comprehensive application in the starch industry. The further research on the physicochemical properties of these *Fritillaria* starches is also going on. The relevant data and information will be reported in succession.

References

Cheetham, N. W. H., & Tao, L. P. (1998). Variation in crystalline type with amylose content in maize starch granules: An X-ray powder diffraction study. *Carbohydrate Polymer*, *36*, 277–284.

Gao, W. Y., Fan, L., & Paek, K. Y. (1999). Ultrastructure of amyloplasts and intracellular transport of old and new scales in *Fritillaria ussuriensis*. *Journal of Plant Biology*, *42*, 117–123.

Krueger, B. R., Knutson, C. A., Inglett, G. E., & Walker, C. E. (1987). A differential canning calorimetry study on the effect of annealing on gelatinization behavior of corn starch. *Journal of Food Science*, *52*, 715–718.

Li, S. L., Chan, S. W., Li, P., Lin, G., Zhou, G. H., Ren, Y. J., et al. (1999). Pre-column derivatization and gas chromatographic determination of alkaloids in bulbs of *Fritillaria*. *Journal of Chromatography A*, *859*, 183–192.

Li, S. L., Li, P., Lin, G., Chan, S. W., & Ho, Y. P. (2000). Simultaneous determination of seven major isosteroidal alkaloids in bulb of *Fritillaria* by gas chromatography. *Journal of Chromatography A*, *873*, 221–228.

Li, S. L., Li, P., & Lin, L. (1999). Existence of 5α-cevenine isosteroidal alkaloids in bulbs of *Fritillaria* L. *Acta Pharmaceutica Sinica*, *34*(11), 842–847.

Li, S. L., Lin, G., Chan, S. W., & Li, P. (2001). Determination of the major isosteroidal in bulbs of *Fritillaria* by high-performance liquid chromatography coupled with evaporative light scattering detection [J]. *Journal of Chromatography A*, *909*, 207–214.

Lin, G., Li, P., Li, S. L., & Chan, S. W. (2001). Chromatographic analysis of *Fritillaria* isosteroidal alkaloids, the active ingredients of Beimu, the antitussive traditional Chinese medicinal herb. *Journal of Chromatography A*, *935*, 321–338.

Nara, S., & Komiy, T. (1983). Studied on the relationship between water-saturated state and crystallinity by the diffraction method for moistened potato starch. *Starch*, *35*, 407–410.

Ruan, H. L., Zhang, Y. H., & Wu, J. Z. (2002). Advances in studies on non-alkaloid constituents of *Fritillaria* L. plants. *Chinese Traditional and Herbal Drugs*, *33*(9), 858–860.

Singh, J., & Singh, N. (2001). Studies on the morphological, thermal and rheological properties of starch separated from some Indian potato cultivars. *Food Chemistry*, *75*, 67–77.

Vasanthan, T., & Bhatty, R. S. (1996). Physicochemical properties of small and large granule starches of waxy, regular, and high amylase barleys. *Cereal Chemistry*, *73*, 199–207.

Yu, S. C., & Xiao, P. G. (1992). The existence of isosteroidal alkaloids in *Fritillaria* L. (Liliaceae) and its taxonomical significance. *Acta Phytotaxonomica Sinica*, *30*(5), 450–459.

Zobel, H. F. (1964). X-ray analysis. In R. L. Whistler (Ed.), *Methods in carbohydrate chemistry* (pp. 109–133). New York: Academic Press.

Zobel, H. F. (1988). Starch crystal transmissions and their industrial importance. *Starch/Stärke*, *40*, 1–7.