Food Sci. Technol. Res., **8** (4), 323–327, 2002

Disintegration Differences in Cooked Potatoes from Three Japanese Cultivars: Comparison of the Properties of Isolated Starch, Degree of Cell Separation with EDTA, and Contents of Calcium and Galacturonic Acid

Chie MATSUURA-ENDO,[1] Akiko OHARA-TAKADA,[1] Hiroaki YAMAUCHI,[1] Yuji MUKASA,[1] Motoyuki MORI,[1] and Ken-ichi ISHIBASHI[2]

[1] *Department of Upland Agriculture, National Agricultural Research Center for Hokkaido Region, Memuro, Hokkaido 082-0071, Japan*
[2] *Department of Bioresource Chemistry, Obihiro University of Agriculture and Veterinary Medicine, Obihiro, Hokkaido 080-8555, Japan*

Received January 15, 2002; Accepted June 26, 2002

Causes of the differences in degree of disintegration after cooking in three potato cultivars with the same starch content, Kita-akari, May Queen, and Hokkaikogane were sought by studying, properties of isolated starches, degree of cell separation in EDTA treatment, and amounts of calcium and galacturonic acid on tubers. The isolated starch of Hokkaikogane had the highest swelling power value and the highest phosphorus content, although this cultivar had the lowest degree of disintegration after cooking of the three cultivars. The degree of cell separation with EDTA, on the other hand, was higher in Kita-akari and lower in Hokkaikogane under conditions without starch swelling. Hokkaikogane had the highest calcium content in the NaCl-insoluble fraction, followed by May Queen and Kita-akari. Furthermore, Hokkaikogane had the highest content of galacturonic acid. These results suggest that factors related to the cohesion of the cells, such as the amounts of calcium and galacturonic acid, are more related to the degree of disintegration after cooking than the properties of the isolated starches in these cultivars.

Keywords: cooked potato, disintegration, isolated starch, cell separation, calcium content, galacturonic acid content

For consumers and processors, the selection of potatoes that have the properties suited for a particular way of cooking or processing is important. The specific gravity of tubers and their degree of disintegration after cooking have often been correlated, particularly within one cultivar (Burton, 1989; Storey & Davies, 1992). Therefore, starch content, which is calculated from the specific gravity, is frequently used in Japan by processing industries and catering institutions, as well as breeding studies as a practical guide for predicting the degree of disintegration after cooking. However, there are instances in which several cultivars differ in their degree of disintegration, even though their starch contents are the same (Ohara *et al*., 1997). We have used three cultivars, Kita-akari, May Queen, and Hokkaikogane, and attempted to determine their properties related to disintegration. Each cultivar exhibited a different degree of disintegration after cooking when they had the same starch content; for example, at 16%, the degree of Kita-akari was "moderate-complete;" that of May Queen was "moderate;" and that of Hokkaikogane was "none." In a previous study, differences in starch distribution within one tuber and tissue structure of a tuber were recognized as possible causes of the difference in degree of disintegration in these three cultivars with the same starch content (Matsuura-Endo *et al*., 2002). Kita-akari, in particular, had a structure in which cells were easily separated by intracellular pressure, which increased by starch gelatinization, and by the breakdown of cell-wall middle lamella that occurred during cooking. Jarvis *et al*. (1992) reported that the starch swelling pressure in cells is great enough to separate potato cells when the middle lamella has been weakened by pectin degradation. In this study, we wanted to understand how the starch and components related to the cohesion of the cells cause differences in their degree of disintegration after cooking among the cultivars. For this purpose, we compared the properties of the isolated starch, the degree of cell separation with EDTA under conditions without starch swelling, and the contents of calcium and galacturonic acid in the tubers.

E-mail: mechie@affrc.go.jp
EDTA, ethylenediamine-*N,N,N′,N′*-tetraacetic acid

Materials and Methods

Potatoes Tubers of three cultivars, Kita-akari, May Queen, and Hokkaikogane, were grown in a field at the Department of Upland Agriculture, harvested in early September 1998, and stored at 4°C and 90–95% relative humidity after curing for two weeks at 15°C. Approximately 1 kg (total weight) of tubers (80 to 120 g each) was used for starch isolation. The starch content of the 1 kg tubers from cultivar Kita-akari, May Queen, and Hokkaikogane was about 15, 15 and 16%, respectively. For other analyses, tubers weighing between 100 and 120 g were selected, and the starch content of each individual tuber was measured using a potato gage (DPG-0.3K, Asahikawa Keiryo, Asahikawa).

Starch isolation and propertie Starches were isolated from approximately 1 kg of tubers of the three cultivars as described by Sabiniano *et al.* (1994). Isolated starches were kept in polyethylene bottles until analyzed. Phosphorus content was measured using the Vanado-Molybdate method described previously (Sabiniano *et al.*, 1994). Swelling power was determined by heating 1000, 250, 250 mg (dry weight) of isolated starch in

50 ml of water at 60, 70 and 80°C (±1°C), respectively, and weighing the swollen sedimented starch, using the method of Yamamoto (1984). The behavior of starch during heating was analyzed using a Brabender Viscograph AM-3 (C.W. Brabender Co., Duisberg, Germany) as previously reported (Sabiniano *et al.*, 1994).

Analysis of the breaking stress and the degree of cell separation on 50℃-EDTA treated tubers The starch content of each individual tuber used in these analyses was 15.5%. The tubers were hand-peeled, cut lengthwise into halves, and treated with/without 0.5% EDTA in a 10 mM Tris-HCl buffer, pH 7.5, containing 0.5% sodium azide at 50°C for 32 h. After treatment, the tubers were washed twice with distilled water and placed in clean distilled water (20°C) for one hour prior to the measurement of their breaking stress. The breaking stress (N/m^2) was determined using a Rheoner (RE-33005, Yamadwen, Tokyo). A cylindrical plunger (3 mm diameter) attached to the instrument was inserted into the treated tubers from the cut surface to a depth of 10 mm at a speed of one mm/s. Nine positions were measured at 10 mm inside from the skin, excluding areas near the eyes. To estimate of the degree of cell separation, treated tubers were cut crosswise (transversely) in the central area between their bud and stem end into slices 1 cm thick. The pieces were then immersed in 25 ml of distilled water in a 50 ml Falcon tube and shaken at 150 rpm for one hour with a reciprocal shaker. The degree of cell separation was estimated by the fresh weight of non-separated tissue that remained in the tube.

Calcium content The starch content of each individual tuber used in this analysis was 15%. Ethanol was added to tuber slices (ca 10 g fresh weight) to a final concentration of 80% (v/v) and a final volume of 30 ml, and the suspension was incubated at 30°C for 18 h and centrifuged. The residue was suspended in 30 ml of distilled water and incubated at 30°C for 18 h and centrifuged. Then, 30 ml of 1 N NaCl was added to the residue, and the suspension was incubated at 30°C for 18 h and centrifuged. Calcium content of this supernatant was called the "1 N NaCl-soluble fraction." Subsequently, 30 ml of 2% acetic acid was added to the residue, and the suspension was incubated at 30°C for 18 h and centrifuged. Finally, 30 ml of 0.6 N HCl was again added to the residue, and the suspension was incubated at 30°C for 18 h and centrifuged. Each supernatant and the final residue were dried, and the calcium content in each fraction was determined by atomic absorption analysis. The calcium contents of the fractions of acetic acid-, HCl-extracted and final residue were pooled and called the "NaCl-insoluble fraction," and the total calcium content was calculated by adding the above fractions.

Galacturonic acid content The starch content of each individual tuber used in this analysis was 15%. Tuber slices (ca 10 g fresh weight) were homogenized with an Ultra-Turrax (T25, IKA WORKS, Selangor, Malaysia) for 5 min in 60 ml of 95% ethanol, and the residue was collected on a filter paper and washed two times with 75% ethanol. Then, 100 ml of 0.5% EDTA (pH 11.5, adjusted with NaOH) was added to the residue and the suspension was incubated at 25°C for 30 min. pH of the suspension was adjusted to 5.0 with acetic acid, and 0.05 g of pectinase (from a mould, Fluka) was added and the mixture was incubated at 30°C for one hour. The mixture was filtered, and the content of galacturonic acid of the filtrate was measured by a carbazole-sulfuric acid method.

Results

Properties of isolated starch The swelling power index of isolated starches from the three cultivars is shown in Fig. 1. The index of Hokkaikogane was higher than that of the other two at the temperature ranges used in this study, especially at 80°C, the value reached 89. Also, Hokkaikogane had the highest content of phosphorus, 1034 ppm, followed in order by Kita-akari and May Queen (Table 1). The starches of Hokkaikogane had different patterns of Brabender viscosity than the other two cultivars (Table 2): the peak viscosity was the highest, and the final viscosity at 92.5°C, was the lowest. Yoshioka *et al.* (1977) reported that the phosphorus content was positively correlated with the peak viscosity of the starch pastes in Japanese cultivars. The peak viscosity of the starch from Hokkaikogane was 1.3-fold higher than that of Kita-akari, a ratio consistent with that of the phosphorus content in this study. These relationships were similar to those in Yoshioka's report (1977).

Breaking stress and degree of cell separation on 50℃-EDTA-treated tubers A previous study (Ohara *et al.*, 1997) showed that Hokkaikogane disintegrated at a much lower degree than the other two cultivars when the starch contents were equal. On the other hand, the isolated starch of Hokkaikogane had the highest swelling power of the three (Fig. 1). We then compared the process of disintegration on tubers of the three cultivars under a condition in which the starches were not gelatinized and not swollen in tuber cells. From the Brabender viscosity patterns (Table 2), the initial pasting temperatures of the three cultivars

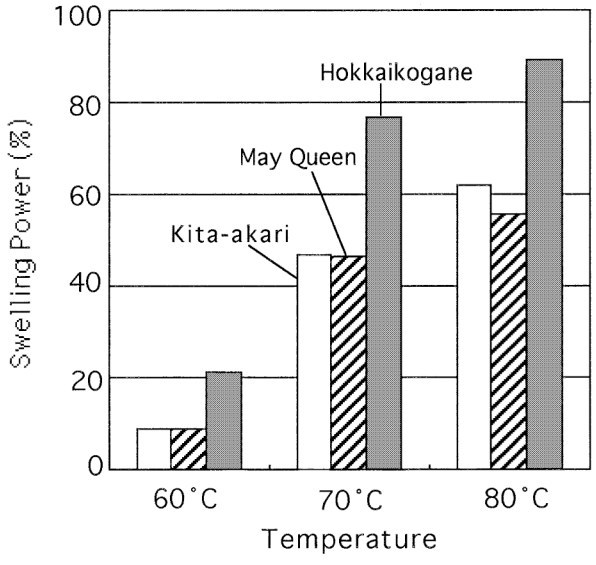

Fig. 1. Swelling power of potato starches from three cultivars. The values are the means of two replicates.

Table 1. Phosphorus content of starches isolated from three potato cultivars.

Cultivar	Phosphorus content	
	(ppm)	(%)
Kita-akari	762	100
May Queen	602	78.9
Hokkaikogane	1034	135.5

Values are the means of two replications.

Table 2. The characteristics of Brabender viscoamylograms of starches isolated from three potato cultivars.

Cultivar	Pasting temp. (°C)	Peak visco. temp.(°C)	Viscosity (BU)		Breakdown (BU)
			Peak	92.5°C	
Kita-akari	64.95	87.01	1345 (100)	485 (100)	860 (100)
May Queen	63.15	76.30	1333 (99.1)	495 (102.1)	838 (97.4)
Hokkaikogane	62.63	67.61	1783 (132.6)	285 (58.8)	1498 (174.2)

Values in parentheses represent the percent of the values ot Kita-akari. Values are the means of two replications.

were in the range of 62–65°C. Therefore, we treated the tubers in a buffer for 32 h at a temperature that did not induce gelatinization of the starch, *i.e.* 50°C. Microscopic observation showed that the starch granules under this condition still exhibited a "Maltese cross" under polarized light (photos not shown), indicating that the granules were ungelatinized birefringent granules (Watson, 1964; Thomas & Atwell, 1997). We also added 0.5% EDTA to the buffer to chelate divalent ions of the tuber tissues. The breaking stress on 50°C-EDTA-treated tubers is shown in Fig. 2. On the incubation at 50°C without EDTA, the value of the breaking stress for Hokkaikogane was the largest, followed in order by May Queen and Kita-akari (Fig. 2-A). These values were similar to those of raw tubers (data not shown), indicating that there were no significant changes in hardiness during the 50°C-incubation. The breaking stress for tubers incubated with 0.5% EDTA, however, decreased up to 1.3–1.5% of those of the buffered control (Fig. 2-B). The values were similar to those of boiled tubers under conditions used in a previous study, which suggests that the 50°C-EDTA treatment induced tissue softening. The order of the values among the cultivars was also the same as that of boiled tubers, the value of Hokkaikogane was the highest, followed by May Queen and Kita-akari. Microscopic observation of tubers

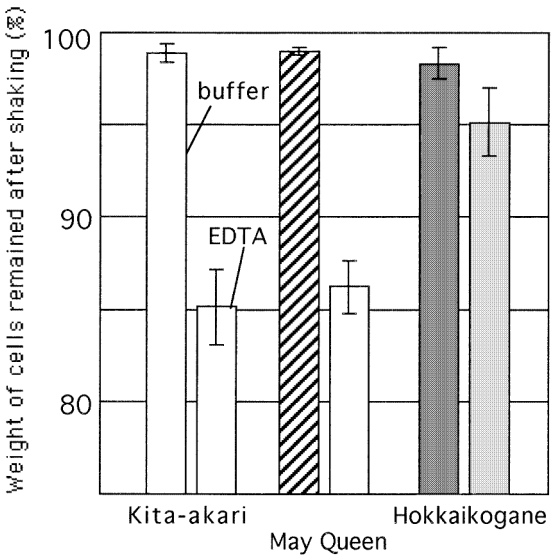

Fig. 3. Analysis of the degree of cell separation on 50°C-EDTA-treated potato tubers. The values represent the percent of fresh weight of the remainder after shaking. Values are the mean ± SE of three independent experiments.

treated with 50°C-EDTA indicated that, although the cell separation occurred in the area of the cut surface to around a 5 mm depth, the cells still had ungelatinized Maltese-cross starches (data not shown). The degree of cell separation in the 50°C treatment was 1–2% for all three cultivars, while those in the 50°C-EDTA treatment were 15% for Kita-akari, 13% for May Queen, and 5% for Hokkaikogane (Fig. 3). These results, also without starch swelling, indicate that Kita-akari and May Queen are more prone to disintegration than Hokkaikogane.

Contents of calcium and galacturonic acid We next measured the contents of calcium and galacturonic acid in tubers,

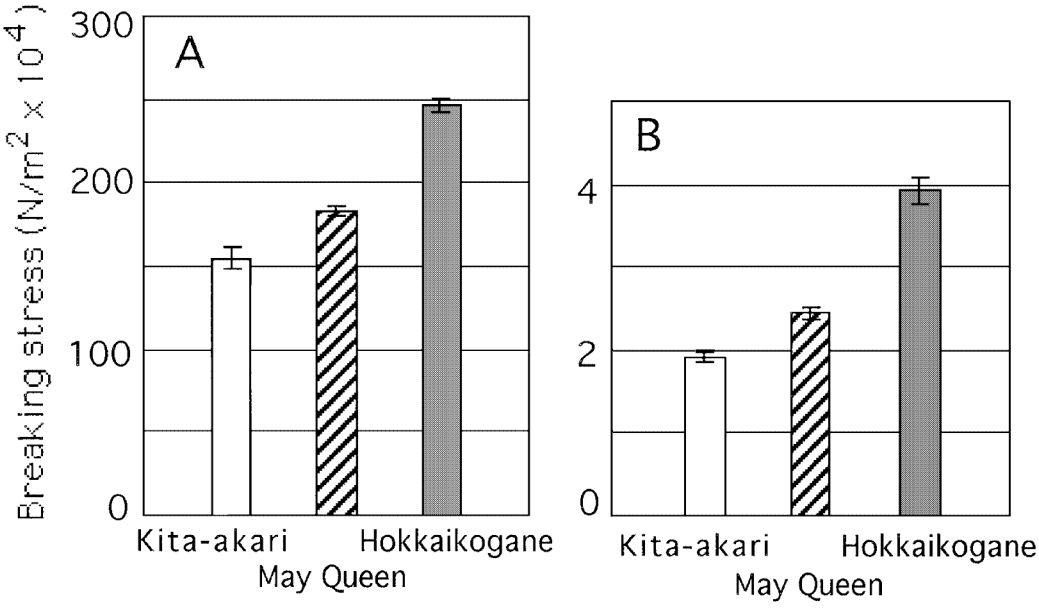

Fig. 2. Breaking stress analysis of 50°C-EDTA-treated potato tubers. A, 50°C for 32 h with EDTA. B, 50°C for 32 h with EDTA. Values are the mean ± SE of three independent experiments.

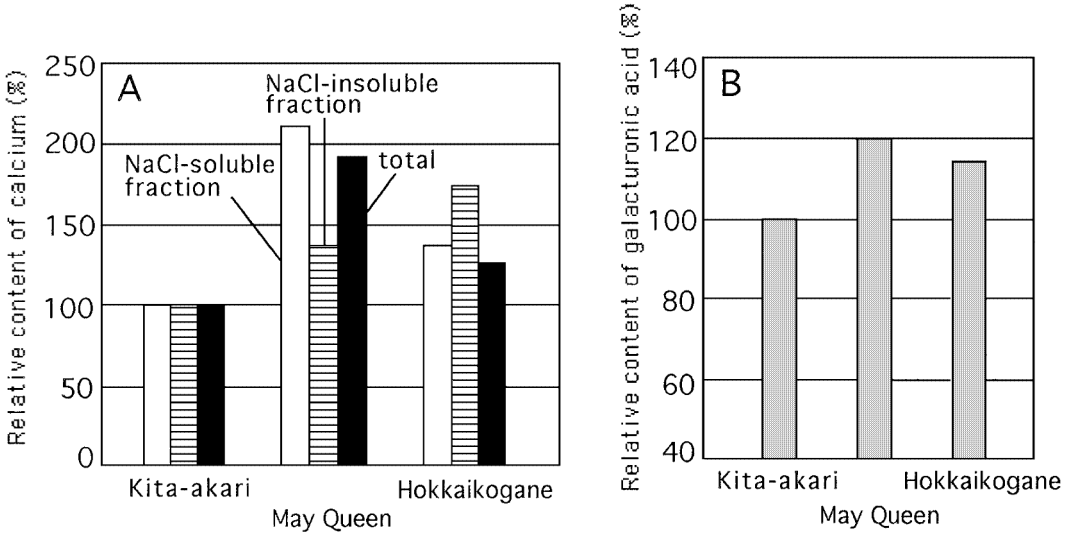

Fig. 4. Contents of calcium and galacturonic acid of potato tubers from the three cultivars. Values are the means of three replicates and represent the percent of the values of Kita-akari. A, the calcium content of raw tubers. One hundred percent of the Kita-akari values of the fractions of NaCl-soluble, insoluble, and the total correspond to 0.24, 0.33, and 1.11 mg/100 g FW, respectively; B, the content of galacturonic acid of raw tubers. One hundred percent of the Kita-akari value corresponds to 336.9 mg/100 g FW.

which are components of the cell-wall middle lamella and are closely related to the texture of cooked potatoes (Hoff, 1973; Burton, 1989; Sato *et al*., 1998). May Queen had the highest calcium content in both total and NaCl-soluble (pectic acid) fraction, followed by Hokkaikogane and Kita-akari (Fig. 4-A). In the NaCl-insoluble fraction, on the other hand, Hokkaikogane was highest in calcium content. The galacturonic acid content of Kita-akari was lower than the others (Fig. 4-B). These results suggest that, of the three cultivars, Kita-akari seems to have the smallest quantities of joint components in the walls of adjacent cells.

Discussion

A previous study suggested that differences in the starch distribution within one tuber and tissue structure may cause differences in the degree of disintegration in cultivars with identical starch content (Matsuura-Endo *et al*., 2002). To understand the effects of starch on disintegration, the starches of three cultivars were isolated and their properties were compared. Hokkaikogane had the highest values of swelling power, phosphorus, and peak viscosity. It has been reported that the phosphorus content in potato starch is positively correlated with the peak viscosity of the starch pastes that were compared among cultivars (Yoshioka *et al*., 1977; Burton, 1989; Sabiniano *et al*., 1994). Microscopic observation indicated that the starches in the cells were gelatinized and occupied the entire cell volume up to about 80°C. Therefore, if the cells of the three cultivars are the same size and structure, contain the same number of starch granules, and have sufficient water to gelatinize the starch, the starch of Hokkaikogane will have the highest internal pressure inside the cell during boiling. Hokkaikogane, however, had the lowest degree of disintegration among the three. These results suggest that the properties of isolated starch itself have less effect on the degree of disintegration after cooking in these cultivars. Thus, the factors relating to the cohesion of the cells were studied next.

The cell walls of potato tubers are largely hemicelluloses and celluloses within a pectic matrix. The cohesion of the cells, particularly, depends upon the jointly shared pectin layer, called the middle lamella. The pectin substances in the cell wall and linking adjacent cells at the middle lamella consist of polymers of esterified galacturonic acid and polysaccharides, and the polygalacturonic acid chains are joined by calcium or magnesium bridges (Jarvis *et al*., 1981; Burton, 1989). Therefore, the potato tubers were treated with EDTA at 50°C in order to chelate the divalent ions in the pectic substance of the wall. The contents of calcium and galacturonic acid in the tubers were also measured. The results showed that the 50°C-EDTA treatment resulted in tissue softening and cell separation without starch swelling. Kita-akari was the most sensitive to the 50°C-EDTA treatment, while Hokkaikogane was the least sensitive (Figs. 2 and 3); Kita-akari also had the smallest amount of calcium and galacturonic acid. These findings suggest that the cohesion of the cells of Kita-akari would be the weakest of the three cultivars. As regards the calcium content of the NaCl-insoluble fraction, Hokkaikogane had the highest value. Earlier reports indicate that the pectin substance, which can be liberated by chelating agents, was retained in the cell wall by the bridges of calcium in the NaCl-insoluble fraction of radish root (Kobayashi *et al*., 1997). Hokkaikogane would have more bridges by calcium in the NaCl-insoluble fraction, namely the polygalacturonic acid chains, than the others.

This study investigated the differences in the properties of the isolated starches of the three cultivars as well as the degree of cell separation without starch swelling and the amounts of calcium and galacturonic acid. Interestingly, the isolated starch of Hokkaikogane had the highest values of swelling power, although this cultivar had the lowest degree of disintegration. This result seems to indicate that factors related to the cohesion of the cells, such as calcium and galacturonic acid, are more related to the degree of disintegration than the properties of the starches. However, many differences in tissue structure were observed, including cell size, grain size of starch, grain number of grains of starch in the cells, and distribution of the starch within the tuber

(Matsuura-Endo *et al*., 2002). These factors could closely relate to the disintegration degree of each cultivar. Therefore, it is necessary to determine how much each factor affects the disintegration, and the effect of interaction between each factor on disintegration. The effect of the pressure from starch swelling in the cells on disintegration might increase as the middle lamella is weakened by pectin degradation during cooking. The changes in these factors during the cooking process should also be investigated. Future studies of these issues might contribute to better processing conditions, selection of more suitable raw materials for various cooking processes, and more effective breeding.

References

Burton, W.G. (1989). "The Potato." Longman Scientific & Technical, New York.

Hoff, J.E. (1973). Chemical and physical basis of texture in horticultural products. *Hort Science*, **8**, 108–110.

Jarvis, M.C., Hall, M.A., Threlfall, D.R. and Friend, J. (1981). The polysaccharide structure of potato cell walls: Chemical fractionation. *Planta*, **152**, 93–100.

Jarvis, M.C., Mackenzie, E. and Duncan, H.J. (1992). The textural analysis of cooked potato. 2. Swelling pressure of starch during gelatinisation. *Potato Res*., **35**, 93–102.

Kobayashi, M., Asaka, T., Matoh, T. and Sekiya, J. (1997). Interactions between pectic polysaccharides and calcium in the cell walls of radish roots. *Plant Cell Physiol*., **38**, (Supplement), 131.

Matsuura-Endo, C., Ohara-Takada, A., Yamauchi, H., Mori, M. and Fujikawa S. (2002). Disintegration differences in cooked potatoes from three Japanese cultivars: Comparison of starch distribution within one tuber and morphology of tissue. *Food Sci. Technol. Res*., **8**, 252–256.

Ohara, A., Kobayashi, A., Matsuura-Endo, C., Yamauchi, H. and Mori, M. (1997). Relationship between the starch content of potato tubers and the disintegration of boiled potato. *Rep. Hokkaido Branch Crop Sci. Soc. Jpn. & Jpn. Soc. Breed*., **38**, 34–35 (in Japanese).

Sabiniano, N.S., Ishibashi, K., Hironaka, K. and Yamamoto, K. (1994). Effect of storage period and temperature on some properties of potato starch. *J. Soc. Agric. Struct*. **25**, 141–146.

Sato, H., Takano, K., Kozima, T., Tanimura, W. and Kamoi, I. (1998). Effect of pectin substance on texture profiles of potatoes with different specific gravity. *Nippon Shokuhin Hozo Kagakukaishi*, **24**, 355–359 (in Japanese).

Storey, R.M.J. and Davies, H.V. (1992). Tuber quality. In "The Potato Crop," 2nd edition, ed. by P. Harris. Chapman & Hall, London. pp. 507–552.

Thomas, D.J. and Atwell, W.A. (1997). "Starches, practical guides for the food industry." Eagan Press, Minnesota.

Watson, S.A. (1964). Determination of starch gelatinization temperature. *Methods Carbohydr. Chem.,* **4**, 240–241.

Yamamoto, K. (1984). Study on property of potato starch and its usage. Ph. D. Thesis, Hokkaido University, Japan (in Japanese).

Yoshioka, S., Sakaguchi, S., Okuyama, Y., Asama, K. and Murakami, N. (1977). Relationship among potato varieties and the quality of starch products. *Res. Bull. Hokkaido Natl. Agric. Exp. Stn.,* **118**, 113–130 (in Japanese).