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Microstructure and Textural Properties of Cell Mass from Cooked Kidney bean and Soybean

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강남콩 고물과 대두 고물의 미세구조와 조직감에 대한 연구

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Abstract

The microstructure, contents of pectic substances and textural properties of cell masses separated from cooked kindey bean and soybean were investigated. Cooked kidney bean could be easily separated to the individual cells, while soybean yielded mixture of individual cells and cell clusters. The break down of the middle lamella was observed, but cell wall was not injured. Separated cells from kidney bean were spherical, while those from soybean were long sack shape as observed in both optical microscope and SEM. Cooked soybean cell mass, which had higher content of pectic substances, showed higher cohesiveness and adhesiveness compared to kidney bean cell mass.

Introduction

The influence of processing and cooking on the cellular structure of dry bean has been studied widely.

Saio and Baba[1] studied the changes in the microstructure of soybeans during storage, and related them to the cooking property and other functional properties of the beans. Soaking in water did not change the microstructure of protein bodies and spherosomes in the intact cells of soybean cotyledone, while remarkable changes, such as bursting of protein bodies and converting of spherosomes into large oil droplets were resulted from heat-steaming and fermentation.[2] Watanabe and Kozuma[3] studied the structure of cooked red bean mash Ann, which is used for Japanese traditional cake. The structure of individual cells separated during Ann process was important for the quality of final product. Starch granules in red bean were swollen during cooking and grinding process, but not solubilized.

Insolubilization of protein appeared to inhibit the gelatinization of strach mechanically.

In the present study, the microstructure of cell masses separated from cooked kidney bean, matured soybean and unripened soybean were observed by using optical and scanning electron microscope. The textural properties of the separated cell mass were compared with the microstructure and the fate of pectic substances of the cotyledones.

Materials and Methods

Materials

Matured soybeans (*Glycine Max*, 1982 crops) and kidney beans (*Phaseolus Vulgaris*, 1983 crops) harvested in Japan were used. Unripened soybeans were purchased from a market in Tokyo.

Methods

Proximate Chemical Analysis: The proximate chemical analysis of the beans and cell masses were made by standard AOAC methods.[4]

Pectic Substances: For the determination of pectic substances, the dry beans were ground in a Willey Mill to make 60 mesh powder. Unripened soybeans were freezed-dried and ground. Twenty percent of water was added to the sample powder and heated at 105°C for 60 minutes in order to increase the extractability of the galacturonic acid. Samples were extracted in 75% ethanol. Chelating treatment was given with 0.5% EDTA·2Na, and then treated with pectinase at 45°C for 3 hours at pH 5. Pectic substances were determined by the m-hydroxydiphenyl determination with spectrophotometer

at 520 nm.[5]

Sample Preparation: Dry beans were soaked in water at room temperature for 20 hours, cooked at 100°C for 60 minutes and then ground. Separated cells were obtained by successive grinding, sieving and washing as shown in Fig. 1.

Texture Measurement: Seven grams of sample were taken from the mass of separated cells and the concentration of sugar was adjusted to 50°Brix. Samples were taken in Tensipresser sample holder and compressed with flat circular probe in two consecutive bites.The textural properties, hardness, cohesiveness, adhesiveness and gumminess, of the mass of separated cells were evaluated by Tensipresser (Model TTD-50x, Taketomo Co. LTD) (chart speed: 60 cm/min., write time; 20 second, load cell; 1 Kg, total speed; 120 mm/min., temperature; 21°C±2).

Microscopical Observation: For the optical microscopic observation, the separated cells from beans were embedded in epon resin and cut with microtome and statined with Schiff's-periodic reagent for polysaccharide, Commassiie brilliant blue for protein, Sudan black B for lipid and ruthenium red for pectic substance.[6] The separated cells were dehydrated with ethanol, coated with gold and observed with scanning electron microscope (SEM) (JSM-20).

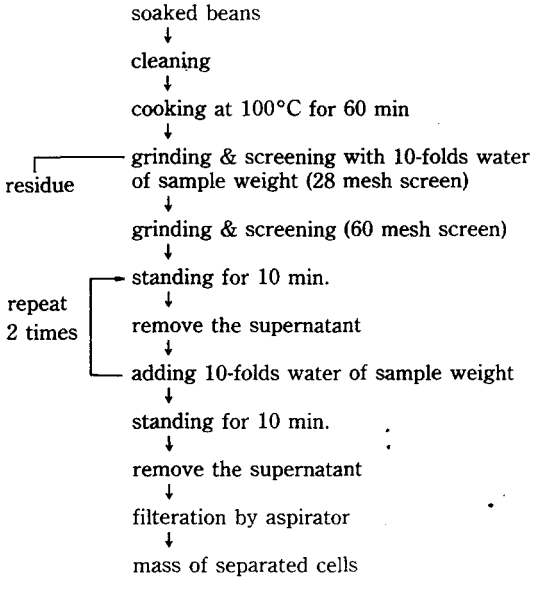


Fig. 1. Flow chart of separated cell mass preparation

Results and Discussion

Chemical composition

Table 1 shows the yield of separated cell mass from cooked beans. Matured soybean showed highest yeild of 75%, whereas kidney bean had the lowest yield. Most of the losses were caused by leaching during washing.

The separated cell masses consisted of 67-74% water, and unripened soybean had higher water content in the cell mass compared to matured beans. The proximate chemical composition of separated cooked cell masses showed that kidney bean contained low fat but high carbohydrate (Table 2). Kidney bean contained comparatively large amount of total pectic substance as shown in Table 3. However, 44% of pectic substances of dry kindey bean

Table 1. Yeilds of separated cell mass from cooked beans

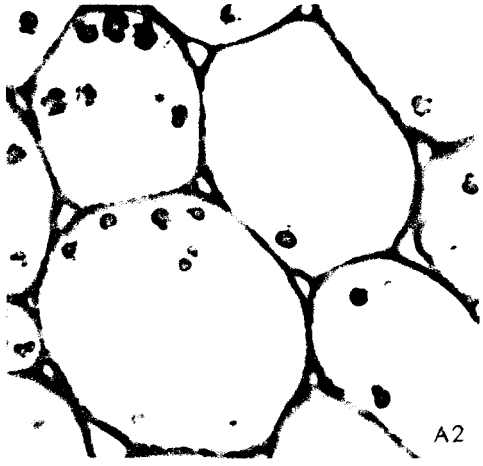
	Mass of separated cells (%)	Residue (%)	Leaching loss (%)
Matured soybean	75.3	3.6	21.1
Unripened soybean	67.9	5.5	26.7
Kidney bean	61.0	15.8	23.2

Table 2. Chemical constitutients of cell mass from cooked beans (dry matter basis)

	Water (%)	Fat (%)	Crude protein (%)	Ash (%)	Carbohydrate (%)
Matured soybean	67.4	27.0	48.5	2.5	22.1
Unripened soybean	73.7	23.9	43.5	2.3	30.3
Kidney bean	68.4	0.5	25.1	0.9	73.5

Table 3. Content of total pectic substances in the fractions of cooked beans

	Mature soybean (%)	Unripened soybean (%)	Kidney bean (%)
Raw bean	3.02	3.16	4.0
Separated cells	2.27	2.33	1.40
Residue	040	0.53	1.77



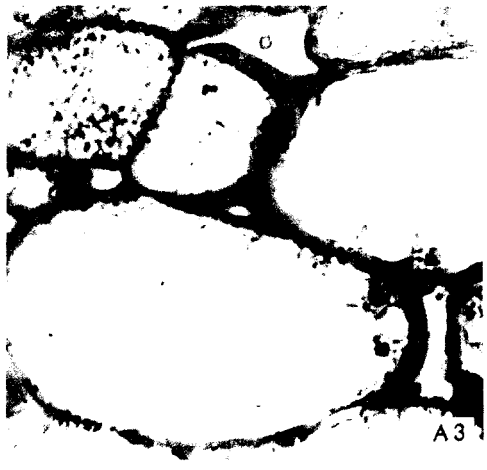
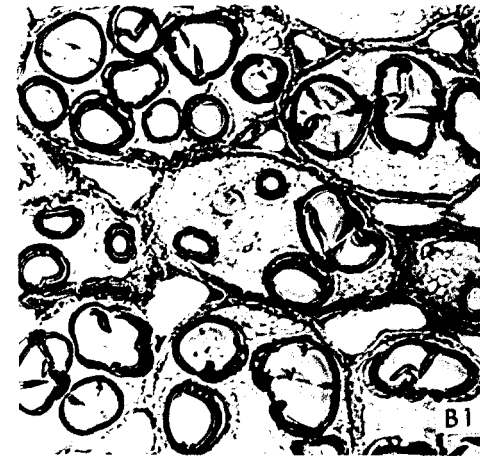
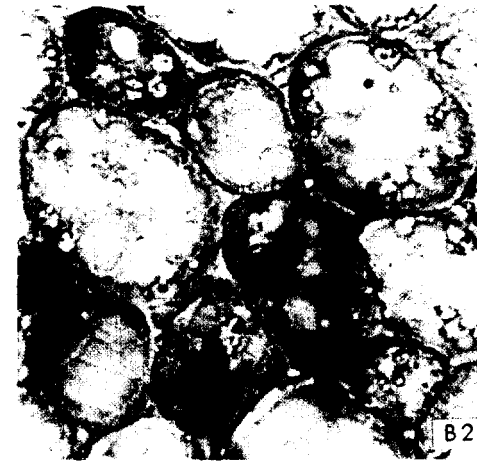


Fig. 2. Microstructure of dry beans stained with Schiff's-Periodic reagent
A1: kindey bean (×400)
A2: unripened soybean (×400)
A3: matured soybean (×400)





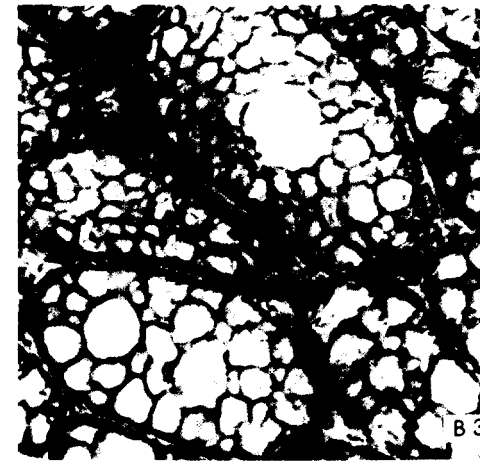
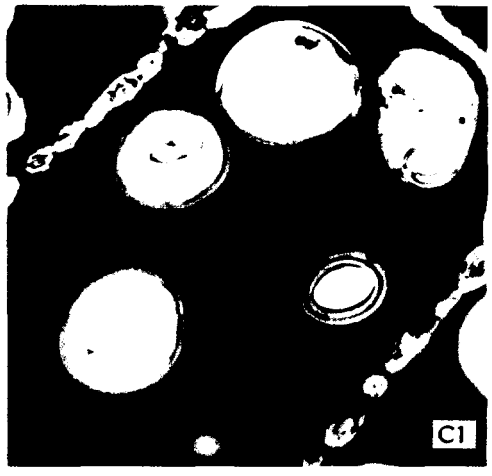
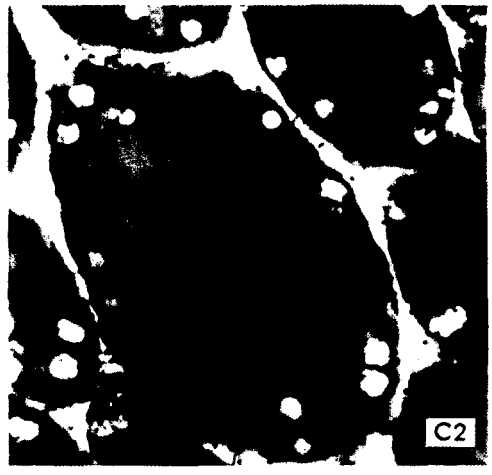


Fig. 3. Microstructure of dry beans stained with ruthenium red
B1: kidney bean (×100)
B2: unripened soybean (×400)
B3: matured soybean (×400)





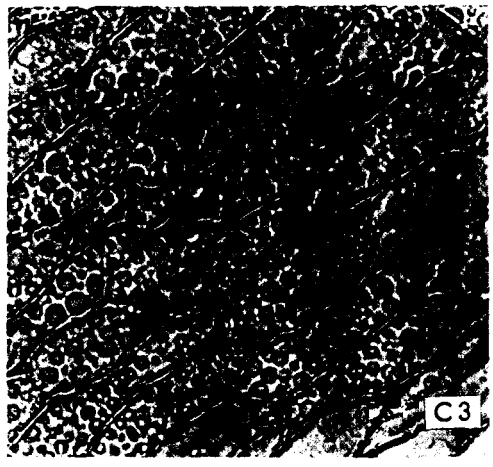
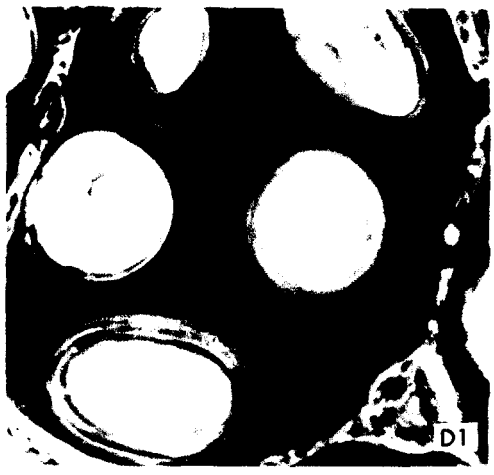
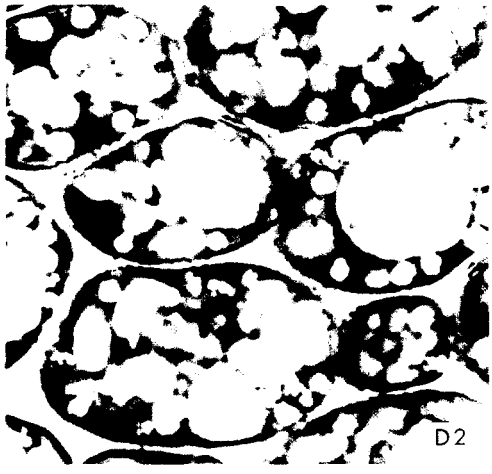


Fig. 4. Microstructure of dry beans stained with coomassiie brilliant bule
C1: kidney bean (×400)
C2: unripened soybean (×400)
C3: matured soybean (×100)





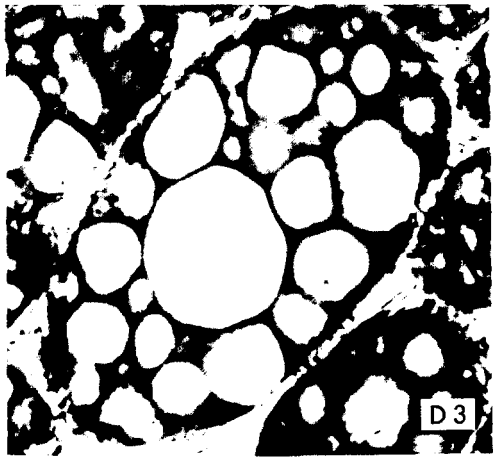
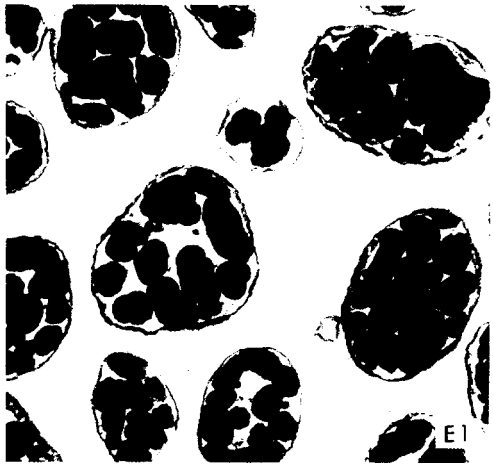


Fig. 5. Microstructure of dry beans stained with sudan black B
D1: kidney bean
D2: Unripened soybean
D3: matured soybean



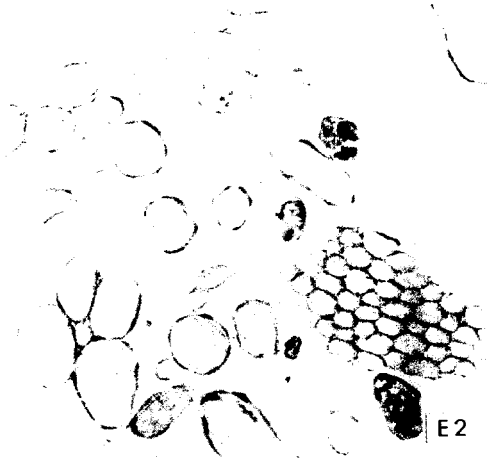
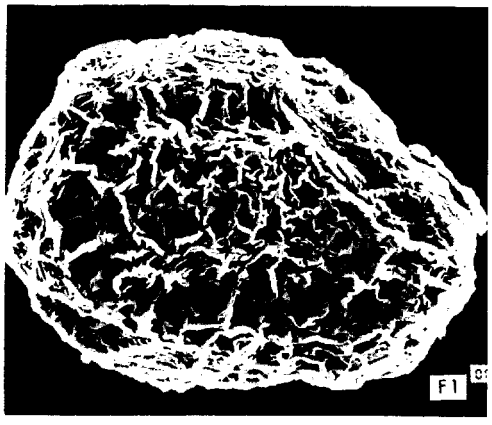


Fig. 6. Microstructure of cell masses separated from cooked beans in PAS staining
E1: kidney bean cell mass (×100)
E2: matured soybean cell mass (×400)
E3: unripened soybean cell mass (×400)



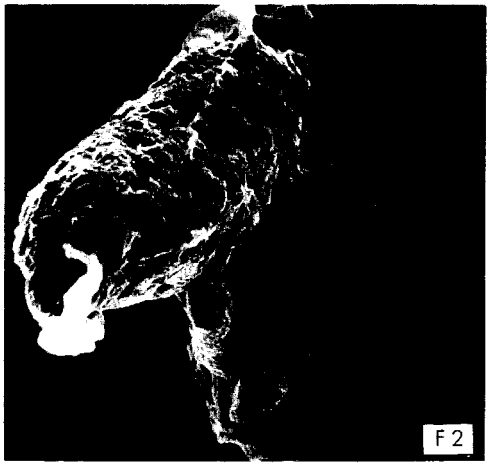


Fig. 7. SEM images of individual cells in the cell mass separated from cooked beans
F1: kidney bean (×1000)
F2: unripened soybean (×1000)
F3: matured soybean (×1000)

were discarded in the residue and 21% were lost as soluble pectin, and only 35% remained in separated cells. In case of soybeans, 74-75% of pectic substances were retained in separated cells.

Microscopic Observation

In Fig. 2, photographs A1, 2, 3 show the microstructure of the dry beans stained with Schiff's-periodic reagent (PAS). Cell wall and starch granules were stained to red. Some starch granules were observed in unripened soybean and their sizes were very small as compared with those in kidney bean. In the matured soybean, most of the starch granules disappered. It agrees with the result of Bils and Howell.[7] In Figures 3,4 and 5, Pictures B1, 2,3 show the microstructure of dry beans stained with ruthenium red, pictures C1,2,3, with coomassiie brilliant blue for protein and pictures D1,2,3, with Sudan black B for lipid, respectively. The middle lamella was stained into weak pink with ruthenium red. This reaction is known to be characteristic for various derivatives of pectic acid. Proteinaceous matrix and protein bodies were stained to violet with coomassiie brilliant blue. Lipid component was stained to dark droplet. These pictures clearly distinguish the structural composition of strach granules, protein bodies, cell wall and middle lamella of the cotyledones. In figure 6, Pictures E1,2,3 show the microstructure of separated cell masses stained with Schiff's-periodic reagent. Cell masses from kidney bean consisted of separated individual cells(E1) formed by the break down of middle lamella, same as observed by Watanabe et al.[8] Soybean cells, both mature (E2) and unripened beans (E3), did not separated well from each other showing many clusters of cells in the separated cell mass.

The large amount of pectic substances in soybean cell masses coincided with the difficulties in separating individual cells. It can be postulated that the cementing effect of pectic substances between cell walls of soybean is stronger compared to that of kidney bean. As shown in Figure 3, the concentration of pectic substance in the middle lamella of kidney bean appeared to be higher than soybean. But substantial portion of pectic substances in kidney bean was leached out as soluble pectin during the process of cooked cell mass making.

Photographs F1,2,3 in Figure 7 show the three dimensional images of the separated individual cells as observed by SEM. It shows that the separated cells from kidney bean has round shape, while those from soybeans have a long sack shape. Picture F3 shows the plasmolized structure between cytoplasm and cell wall.

Textural Properties

Table 4 shows the textural properties of the cell masses separated from cooked beans. Kidney bean shoed higher hardness, but lower cohesiveness, adhesiveness and gumminess compared to soybeans. The cooked cell mass from matured soybean had higher adhesiveness and higher gumminess than those from unripened soybean. Addition of sugar to 50°Brix increased the hardness of cell mass. The cohesiveness, adhesiveness and gumminess of matured soybean cell mass decreased by the addition of sugar, while those of kidney ban and unripened soybean increased. It would be significant to compare the textural properties of kidney bean and matured soybean, because these cell masses contained similar amount of water, which exerts a great influence on textural properties. It was interesting to note that the cohesiveness and

Table 4. Textural property of mass of separated cooked cells

				Sugar added (50°Brix)		
	Kidney bean	Unripened soybean	Matured soybean	Kidney bean	u.s.b.	m.s.b.
Hardness (Kg. weight)	1.18	0.94	0.81	2.11	1.45	1.40
Cohesiveness	0.09	0.83	0.81	0.13	1.10	0.73
Adhesiveness (Kg/sec)	0	1.69	2.40	0.04	2.27	1.91
Gumminess (Kg. weight)	0.11	0.78	1.36	0.27	1.60	1.02

u.s.b.: unripened soybean m.s.b.: matured soybean

adhesiveness was much greater in matured soybean cell mass, which contained higher concentration of pectic substances.

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요 약

강남콩과 대두를 삶아 분리한 고물의 미세구조를 광학 현미경 및 SEM으로 조사하고 그 화학조성과 조직감 특성을 비교하였다. 강남콩 고물의 경우 각각의 세포가 잘 분리되었으나 대두 고물은 여러개의 세포가 결합된 덩어리가 많이 포함되어 있었다. SEM 사진에 의하면 강남콩 고물을 구성하는 세포는 구형이었으나 대두 고물의 구성세포는 긴 자루 형태를 가지고 있었다. 펙틴질을 비교적 많이 함유하는 대두 고물의 경우 높은 접착성과 응집성을 나타내었다.

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