Enhancement of Sweet Components in Vegetable Soybean Seeds: Starch Degradation During Cooking Enhance Flavor of Immature Seeds

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Introduction

The most important taste of boiled edamame is sweetness (Masuda et al, 1988). The components responsible for sweetness of raw vegetable soybean seeds are sugars and free alanine, mainly sucrose (Masuda, 1991). Thus, sucrose level in edamame seeds in retail is an important factor of the quality as in case of sweet corns, melons and oranges, etc. Decline of sucrose during three day's storage at ambient temperature is unavoidable, even if the harvest pods are wrapped with less-transparent (somewhat anaerobic condition) or micro-perforated films (Masuda, 2000).

In sweet corns, the enhancement of sweet components in kernels is made successfully by reduction of starch synthesis activities and increase in sucrose accumulation. This strategy is a considerable method in sucrose enhancement of immature soybean seeds. Carbohydrate contents of mature seeds were reported in various genotypes and at various cultural conditions, but there are few data when and how much sucrose and starch accumulate in these developing seeds. Also, carbohydrate composition at mature did not reflect the compositional values of developing seeds.

The first aim of this paper was to reveal compositional changes of soluble sugars and starch accumulating during development at normal field condition of cultivars used for traditional soy foods, and categorize some seeds based on sucrose and starch accumulation. The second was to answer other possibility of enrich methods of sweetness. The course of studies on factors responsible delicious tastes of boiled immature Tanbakuro seeds; we found sugars formed during the boiling.

Methods

1. Cultivation and sampling

Soybeans were grown at the fields of Natl. Inst. Agrobiol. Resour. and Natl. Agric. Research Center (Tsukuba, Japan) using standard cultural practice and fertilized 5-7 kg N, 8-12 kg P, 10-15 kg K/ 10 a before sowing. Seeds were sown on May or June 1990, 1991,1992,1998 and June 1999 (Kim, Kikyo 1 and Sayakaori). Five to ten plants with pods were harvested from early green stages to mature stages of developing seeds. Pods were picked from the plants in the laboratory, and pod walls were removed. The largest number group of the seeds separated at 0.05 g each was selected for the sample and submerged into liquid nitrogen, kept at -80 ºC until analysis.

2. Component determination

The frozen seeds (about 2 g FW) were added to ten volumes of 80% (v/v) ethanol. Immediately after the addition, the mixtures were homogenized for 2 min with a Polytron homogenizer twice. The centrifuged homogenate was collected. Before HPLC analyses insoluble materials in the extracts were removed centrifuging and passed through a membrane of 10,000 nominal molecular weight limits. Sugar contents were determined by a modified procedure of previously reported HPLC method (Masuda et al., 1996). For the individual determination of pinitol and D-fructose, the separation column was replaced by a

Dionex MA-1 (4.0 x 250 mm I. D.).

The residues of sugar extractions were used to determination of starch contents. For starch determination, the centrifuged precipitates were mixed with water, and then autoclaved for 20 min. Starches were assayed using an F-Kit glucose determination (Boehringer Mannheim).

Results

1. Explore of high sucrose or starch immature seeds among domestic varieties in Japan

Developmental changes in carbohydrate composition were analyzed during the full developing periods in 30 genotypes and partial in 18 varieties. These seeds categorized to four types based on the balance between soluble sucrose and insoluble starch during the development (Tab. 1). Since accumulation patterns of two carbohydrates presents carbohydrate economy of developing seeds. Developing seeds of Enrei, typical cultivars for tofu, contained sucrose, stachyose, raffinose, verbascose, pinitol, myo-inositol, fructose, glucose, galacto-pinitols and starch. Total carbohydrates were increasing at early stages, but after 32 DAF (day after flowering) decreasing (Fig. 1). The maximum of the total carbohydrates, sucrose and starch were 5.0, 2.7and 2.7g/100g FW at 52, 52 and 46 DAF, respectively except mature seeds. Starch declined rapidly prior to the rapid decrease of water content of seeds. Instead of starch, stachyose increased during the same periods.

Seeds of Tamasudare, a typical edamame with green pods, contained 3.3g/ 100g FW sucrose at 26 DAF. Chakaori, its origin seemed as same as Dadachamame, showed that the maximum of the total, sucrose and starch were 7.6, 4.5, 3.3 g/ 100gFW at 32, 32 and 41 DAF, respectively. The total carbohydrates and sucrose of these seeds were the highest values among the analyzed soybean seeds. However, immature Tanbakuro seeds have been known as its good taste, the sucrose content of the edamame stage was 2.0g/ 100gFW. The seed size is largest but needs a longer developing period than Enrei or edamame seeds.

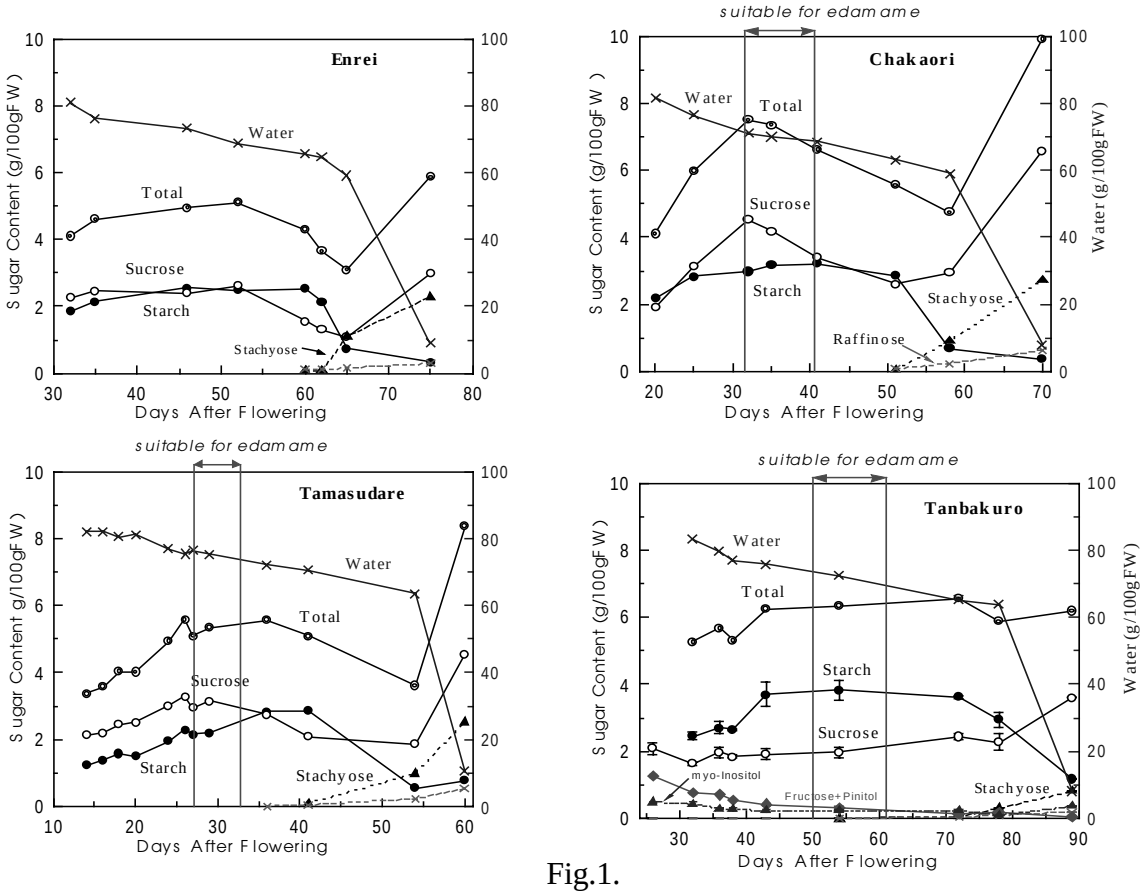

Fig.1.

Changes in carbohydrate and water content of developing soybean seeds suitable for

edamame. Enrei is grain-type.

Table 1. Classification of soybean cultivars on carbohydrate accumulation patterns in developing seeds.

Carbohydrates	Cultivars
Sucrose > Starch	Chakaori, Murasaki-dadacha, Kikyo, Takihime, Tamasudare, Kim, Kokeshijiro, Saikai 20, Ichigou-wase
Sucrose = Starch	Enrei, Fujimijiro, Tousan 90
Sucrose < Starch	Sayakaori, Kuro-Satou, Satou-irazu, Natto-shoryu Satou-daizu (Akita), Satou-daizu (Ishioka)
Sucrose < Starch	Tanokuro, Koito-zairai, Koguro-daizu, kouji-shirazu
Late Sucrose	Mochi-daizu, Tamahikari, Akasaya-daizu, Oogata-aomame
Increasing	Tsutsueao, Akagishita, Akiyoshi-kurakake-mame, Asomusume

2. Alternative way of enhancement of sweet components

Boiled immature seeds of Tanbakuro have delicious taste. We found two factors that were different from other vegetable soybean seeds. The first is softer texture of the boiled seeds than those of other varieties. The hardness of a boiled cotyledon was measured as the maximum force of sticking by a plunger 2.5mm in diameter. Hardness of Tanbakuro was 1.04±0.40 N/m^2 while that of Chamame purchased from the market was 9.76±2.62 N/m^2. Perception of taste components in hard texture foods is weaker than that of soft foods (Matsumoto & Kazama, 1965).

The second is that boiled seeds contained newly formed sugars, maltose and very low level of maltotriose. ‘Hyoukei kuro 3 gou’ seeds at 53DAF produced a reducing sugar 0.9g /100gFW at maximum from starch after pods boiling for 6 min while remaining sucrose was 1.6g /100gFW.

Comparison of maltose formation of developing soybean seeds was done on boiling of the frozen stock seeds. Maltose production of frozen Tanbakuro seeds for 60 sec boiling was equal to that of the raw seeds within pod for 6 min boiling. While developing soybean seeds showed maltose formation 0.3-1.5g /100gFW after boiling, there were some developing seeds with a few amount of maltose production among early grown varieties. Interestingly edamame varieties containing high sucrose belonged to low maltose production seeds after boiling, though starch quantity was enough to maltose formation.

Discussion

Sweet corn kernels contain large amounts of starch, whereas immature soybean seeds have less than 4g /100g FW starch at maximum. The strategy of simply reduction of starch biosynthesis might be difficult to enhance sucrose accumulation in soybean seeds. Sucrose accumulation in developing seeds showed critical roles in carbon supplies to respiration and biosynthesis of lipid, protein and other components. After harvest, sucrose supply from leaves or stems is stopped and decrease of sucrose was inevitable except under low temperature. In addition, seed boiling may result in a release of sucrose. Sucrose levels in harvest and 3 day' stored and boiled seeds of Chakaori are 4.5g and 2.2g /100gFW, respectively. Reduction of sucrose content between raw and boiled edamame was beyond 50%. Edamame contained sucrose below than 1.8g /100g showed bad sensory scores (Masuda et al, 1988). Cultivars with more than 4g /100gFW sucrose at immature green seeds are suitable materials for breeding of edamame.

Reducing sugar newly formed during seed boiling shows the sweetness strength of 0.4 times of sucrose, but more refreshing aftertaste than sucrose. Boiled edamame seeds of Tanbakuro including 0.9g /100gFW reducing sugar in addition to sucrose are more delicious than the seeds only of sucrose.

Conclusions

1. The retrieve of high sucrose containing seeds among domestic varieties showed Chakaori is the richest variety in sucrose and total carbohydrates.
2. The results indicated that the little possibility of enlargement of sucrose pool in developing seeds by means of simply reduction of starch biosynthesis.
3. Reducing sugar formation during seed boiling is a new clue for increasing sweet components.

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