Properties of Tofus and Soy Milks Prepared from Soybeans Having Different Subunits of Glycinin

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The contribution of soybean protein to the physical properties of tofu, a product manufactured by curdling soy milk with coagulants such as calcium or magnesium chloride, was studied by comparing the properties of soy milk prepared from soybeans with different subunits (I, IIa, and IIb) of glycinin with amino acid residues deleted. The breaking stress value of the tofu curds prepared from soybeans having group I was higher than those without group I. The soy milks having group I contained more protein particles and showed more sensitivity to calcium and magnesium ions than those without group I. The amounts of glycinin and protein particles were higher in the soy milks having group I than those in the soy milks without group I. To elucidate the influence of each group on the breaking stress, the glycinin content was adjusted to an identical level in soy milks having each group. Among the tofu curds from three groups, their order of hardness according to their breaking stress was IIa, IIb, and I. The order of particle content among these soy milks was also IIa, IIb, and I. Therefore, the results suggested that the breaking stress value of the tofu curd is dependent upon the number of protein particles in the soy milk and that the number of the particles is determined by the proportion and structure of glycinin in the soybean.

Keywords: *Soybean; tofu; soy milk; glycinin; coagulation; breaking stress*

INTRODUCTION

Soybean foods, such as soy milk and tofu, are popular in some Asian countries. The quality of tofu, a product manufactured by curdling soy milk with calcium or magnesium chloride, depends largely on its physical properties. The protein in soy milk plays an important role in the physical properties of tofu. Soybeans for tofu manufacturing contain about 35% protein, which comprises 40% glycinin and 30% β-conglycinin.

It is known that the hardness of tofu curd is influenced by the ratio of glycinin to β-conglycinin (Saio et al., 1969). Glycinin consists of acidic (A) and basic (B) subunits, which are linked by a single disulfide bond, and β-conglycinin consists of α, α', and β subunits (Mori et al., 1981; Staswick et al., 1984). Before processing in biosynthesis, glycinin is a single peptide in which A and B subunits are linked to each other (Barton et al., 1982). The peptide has five genetic variants, which are named as A1aB1b, A2B1a, A1bB2, A5A4B3, and A3B4 (Nielsen, 1985). At present, glycinin is classified into group I (A1aB1b, A2B1a, and A1bB2) and group II (A5A4B3 and A3B4) based on homology in subunit sequences (Nielsen et al., 1989). Group II is further classified into two subgroups named as IIa (A5A4B3) and IIb (A3B4) (Yagasaki et al., 1996). Three groups (I, IIa, and IIb) differ especially in the carboxyl terminal end of the acidic subunit. In amino acid sequences of groups I and IIb, about 60 and 30 amino acid residues are deleted from group IIa, respectively (Nielsen et al., 1989). The physical property of tofu curds prepared from soy milks containing various groups of a glycinin subunit has not yet been studied.

As tofu is prepared from soy milk, the physical property of tofu curd should depend on the properties of the soy milk. About 50% of soy milk proteins have been found to be protein particles (Ono et al., 1991), which formed the core of tofu curd on coagulation with calcium chloride (Ono et al., 1993). In this paper, to elucidate factors affecting physical properties of tofu curd, we report the relationship between the hardness of tofu curds and the properties of soy milk prepared from soybeans having different subunits of glycinin.

MATERIALS AND METHODS

Materials. Soybeans (*Glycine max* (L.) Merr.) used in this study include eight lines, which differ in glycinin content or in their class of subunits. One line has a low content of glycinin, and the others have the following types of glycinin: group I; group IIa; group IIb; group I and IIa; group I and IIb; group IIa and IIb; and group I, IIa, and IIb (Yagasaki et al., 1997). These soybeans were breed by backcrossing between Tamahomare, a recurrent parent lacking a group IIa subunit, and a line having only the group IIa subunit (Yagasaki et al., 1996). Backcrossing was done for two generations. All seeds were harvested in 1997−1998 and were stored at 5 °C until used.

All of the chemicals were the highest purity available and were used without further purification.

Preparation of Tofu Curd. Soybeans were soaked in deionized water for 20 h at 4 °C. The swollen beans were ground into homogenates with deionized water using an Oster blender (Oster Co, Milwaukee, WI). The homogenates were

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heated for 5 min at 97 °C and filtered through cotton cloth. The filtrates of the soy milk were cooled to 10 °C. A magnesium chloride solution (50 g of $MgCl_2 \cdot 6H_2O$ + 5.0 g of NaCl/200 mL of H_2O) was added to the soy milk, which was poured into a polypropylene vessel and heated for 45 min at 85 °C to prepare the tofu curd. The curd was cooled and kept at 10 °C.

Measurement of Tofu Curd Hardness. A compression test was carried out at a compression rate of 6.0 cm/min using a Sun Rheo Meter model CR-200D (Sun Scientific Co. Ltd., Tokyo, Japan) with a cylindrical plunger of 1 cm diameter. The breaking stress of the tofu curd was expressed by means of seven measurements per one sample.

Determination of Soybean Components. The protein, lipid, ash, and carbohydrate contents of the soybean were determined by using a near-infrared spectrophotometer (Infra Alyzer 500, BRAN LUBBE, Hamburg, German).

Determination of Phytate. Phytate content was measured by the method of Makower (1970) with some modifications. Two milliliters of soy milk was mixed with 2.0 mL of 6% trichloroacetic acid. The mixture was incubated for 20 min at 20 °C and centrifuged at 1800*g* for 15 min. Two milliliters of the supernatant was mixed with 6.0 mL of an iron chloride solution ((2.0 g of $FeCl_3 \cdot H_2O$ + 16.3 mL of concentrated HCl)/1 L of H_2O). The mixture was incubated for 30 min at 97 °C, cooled at room temperature, and then centrifuged at 1800*g* for 15 min. The precipitate was dispersed in 2.0 mL of washing solution (0.6% HCl + 2.5% Na_2SO_4) and then centrifuged at 3000*g* for 15 min. The precipitate was further dispersed in 2.0 mL of 0.3 N NaOH and centrifuged at 1800*g* for 15 min. The precipitate was dissolved in 3.0 mL of 0.3 N HCl, and the volume was adjusted to 10.0 mL with deionized water. The iron content was measured colorimetrically at 510 nm based upon *o*-phenanthroline. It was assigned one phytate molecule linked with four iron molecules, and the weight of phytate could be calculated numerically as 2.98 times the weight of iron.

Preparation of Raw Soy Milk and Soy Milk. Soybeans were soaked in deionized water for 20 h at 4 °C. The swollen beans were ground into homogenates with deionized water using an Oster blender (Oster Co.). The homogenates were filtered through a defatted cotton sheet, and the filtrates were designated as raw soy milk containing 11.5% solids. The raw soy milk was heated in boiling water for 5 min and was designated as soy milk containing 11.5% solids.

Purification of Each Group Protein. After each glycinin was prepared from soybean having each subunit by the method of Thanh et al. (1975), group I, IIa, or IIb protein was purified from the glycinin by using Con A Sepharose (Pharmacia, Uppsala, Sweden).

Preparation of Raw Group−Protein Soy Milk and Group−Protein Soy Milk. The raw soy milk containing 8.5% solids was prepared from soybeans having a low content of glycinin and is designated as raw low group−protein soy milk. Group I, IIa, or IIb protein was added into raw low group−protein soy milk to prepare 10.5% solids. The raw group−protein soy milks were heated in boiling water for 5 min and designated as group−protein soy milks. The soy milk containing group I, IIa, or IIb has the same glycinin content and was designated as I group−protein soy milk, IIa group−protein soy milk, or IIb group−protein soy milk, respectively. These group−protein soy milks were used to investigate the distribution of the protein particles.

Preparation of Protein Particles. Protein fractions, which differ in the size of protein particles, were prepared from raw soy milk, soy milk, raw group−protein soy milk, and group−protein soy milk by the method of differential centrifugation (Ono et al., 1991). Protein particles of more than 100 nm in diameter were obtained in pellet form by centrifugation of raw soy milk, soy milk, raw group−protein soy milk, and group−protein soy milk at 32500*g* for 30 min. Then protein particles of 40−100 nm and less than 40 nm in diameter were obtained in pellet form and supernatant, respectively, by centrifugation of the preceding supernatant at 156500*g* for 30 min. The particles of more than 100 nm, 40−100 nm, and less than 40 nm in diameter were named as large protein particles, medium protein particles, and supernatant protein, respectively. These contents were expressed by means of five measurements per one sample.

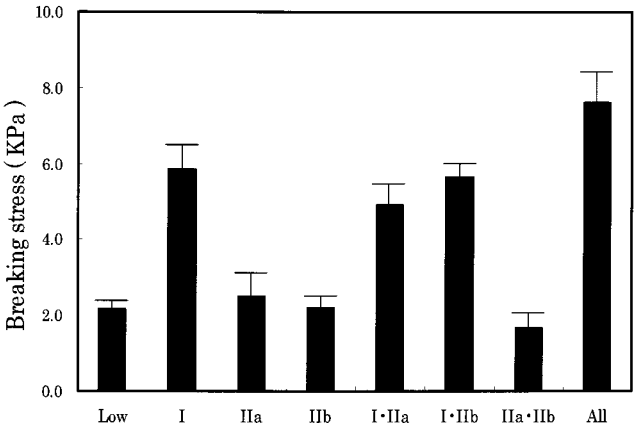

Figure 1. Breaking stress of tofu curds containing 11.5% solids prepared from soybeans having different subunits of glycinin. Values are means ± standard deviation, $n = 7$ per group. Tofu curds were prepared by adding 1.9 mL of magnesium chloride solution (50 g of $MgCl_2 \cdot 6H_2O$ + 5.0 g of NaCl/200 mL of H_2O) from eight soybean lines having a low content of glycinin (Low), group I (I), group IIa (IIa), group IIb (IIb), group I and IIa (I·IIa), group I and IIb (I·IIb), group IIa and IIb (IIa·IIb), and group I, IIa, and IIb (All).

Measurement of Protein Solubility. Measurement of protein solubility was performed according to the method of Ono et al. (1993). The protein solubility of soy milk in the presence of calcium or magnesium chloride was determined by measuring the protein content of the supernatants after centrifugation at 1800*g* for 5 min. The pH of soy milk was checked after the addition of calcium or magnesium chloride.

Determination of Protein. The protein content was measured by the method of Bradford (1976), after lipid had been removed in a procedure described below. Samples were mixed with the same volume of 2 N NaOH, incubated for 30 min at 60 °C, and centrifuged at 8000*g* for 30 min. The medium layer was neutralized with a 0.1 M phosphate buffer (pH 6.0) and used for the measurement of protein.

Electrophoresis. SDS−polyacrylamide gel electrophoresis (SDS−PAGE) was carried out in a vertical slab gel of 1 mm thickness, using an alkaline discontinuous buffer system (Laemmli, 1970). Coomassie brilliant blue G-250 was used for protein staining by the method of Blakesley and Boezi (1977). After electrophoresis, gels were immersed in a staining solution containing 12% trichloroacetic acid for 12 h and then destained with water.

Densitometric analysis of the stained gels was carried out with a BIO-RAD Multi-Analyst instrument (BIO-RAD, Hercules, CA).

RESULTS AND DISCUSSION

Breaking Stress of Various Tofu Curds. Tofu curds were prepared from eight soybean lines that differ in their class of subunits. The breaking stress of these tofu curds was measured and is shown in Figure 1. The breaking stress values of tofu curds prepared from soybeans having a low content of glycinin, group I, group IIa, group IIb, group I and IIa, group I and IIb, group IIa and IIb, and group I, IIa, and IIb were 2160, 5840, 2500, 2220, 4910, 5650, 1670, and 7610 Pa, respectively. The breaking stress values of tofu curds prepared from soybeans having group I were twice the value of those without group I. These values without group I were similar to those having a low content of glycinin.

Compositions of Soybeans and Soy Milks. Compositions of soybeans and soy milks were analyzed since

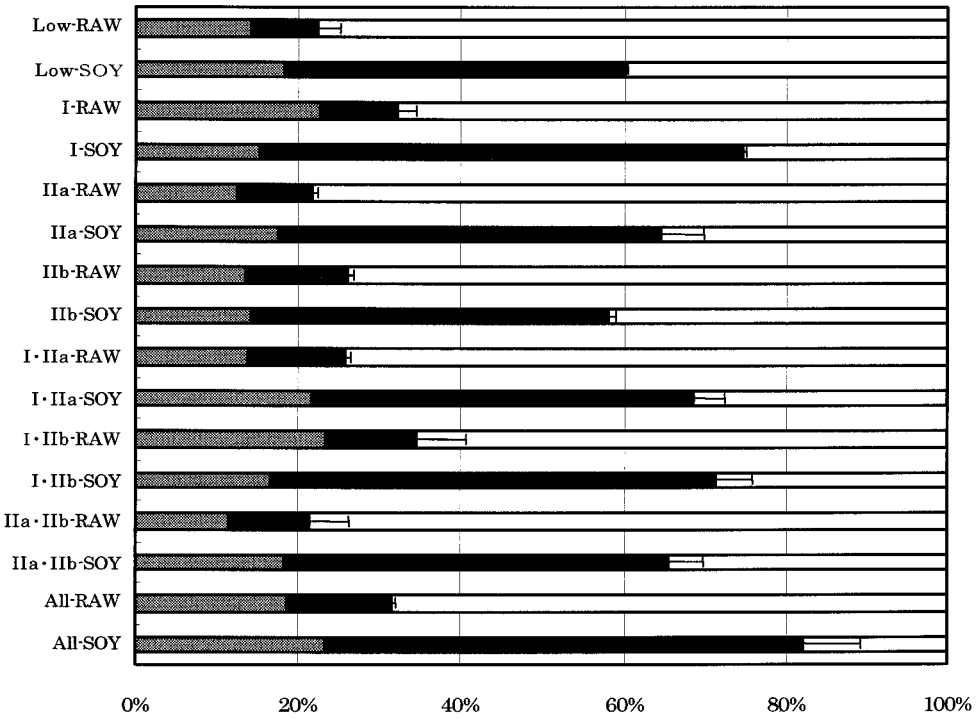

Figure 2. Distribution of the protein particles obtained by differential centrifugation from the raw soy milks (RAW) and soy milks (SOY) prepared from soybeans having different subunits of glycinin. The particles of more than 100, 40−100, and less than 40 nm in diameter were large protein particle (gray bars), medium protein particle (black bars), and supernatant protein (white bars), respectively. Values are means ± standard deviation (sum of large and medium protein particles), $n = 5$ per group. The raw soy milks and soy milks containing 11.5% solids were prepared from eight soybean lines having a low content of glycinin (Low), group I (I), group IIa (IIa), group IIb (IIb), group I and IIa (I·IIa), group I and IIb (I·IIb), group IIa and IIb (IIa•IIb), and group I, IIa and IIb (All).

Table 1. Compositions of Soybean and Soy Milk Having Different Subunits of Glycinin[a]

	in soybean				in soy milk
group	protein (%)	lipid (%)	carbohydrate (%)	ash (%)	phytate (mg/g of protein)
Low	37.57	17.02	40.18	5.23	24.0
I	38.13	18.36	38.24	5.27	24.6
IIa	40.64	16.17	38.22	4.97	26.1
IIb	35.36	19.34	40.20	5.10	23.7
I·IIa	39.46	17.74	37.61	5.19	26.5
I·IIb	38.47	17.46	39.12	4.95	25.1
IIa·IIb	38.57	17.52	39.18	4.73	24.8
All	36.47	18.98	39.53	5.02	27.1

[a] The group Low, I, IIa, IIb, I·IIa, I·IIb, IIa·IIb, and All have low content glycinin,group I, group IIa, group IIb, group I and IIa, group I and IIb, group IIa and IIb, and group I, IIa, and IIb.

the breaking stress values of the tofu curds were influenced by the contents of the components. The protein, lipid, carbohydrate, and ash contents of soybeans having different subunits of glycinin and phytate contents of the soy milks were nearly similar to the soybeans or soy milks shown in Table 1. The protein content of soybeans having a low content of glycinin (Low) was almost the same as that of soybeans having the group I, IIa, and IIb subunits (All). The soybean Low contains a larger amount of β-conglycinin and a low content of glycinin. On the other hand, the soybean All contains a larger amount of glycinin. It is known that phytate in soy milk induces a decrease in pH with the addition of calcium (Ono et al., 1993; Tezuka et al., 1995). Therefore, the phytate content of soy milk affects the breaking stress values of tofu curds. The phytate content among soy milks with different subunits of glycinin showed similar values. These results indicated that the differences in the breaking stress among these tofu curds are due to factors other than the content of each component in the soy milks.

Distribution of the Protein Particles from Various Soy Milks. The physical property of tofu curd depends on the properties of the soy milk. One-half of soy milk protein is in particles more than 40 nm in diameter (Ono et al., 1991), which form the core of tofu curd (Ono et al., 1993). The distribution of the protein particles in raw soy milks and soy milks prepared from soybeans having different subunits of glycinin is shown in Figure 2. Apparently, the proportions of medium protein particles in the soy milks were higher than the respective raw soy milks in all cases. It is noted that the soy milks containing group I had higher proportions of protein particles (large plus medium sized) than those without group I. The breaking stress values of the tofu curds containing group I were already shown to be higher than those from soy milks without group I (Figure 1). This fact suggests a possibility that the hardness of a tofu curd is affected by the proportion of protein particles, which seems to be concerned with the core formation of the tofu curd (Ono et al., 1991).

The subunit compositions of the centrifugal fractions of soy milks prepared from various soybeans were calculated by SDS−PAGE and densitometric analysis. The proportion of protein subunits in these fractions, including large and medium protein particles and supernatant protein, is shown in Figure 3. The soy milks containing group I had larger amounts of glycinin than those without group I (Figure 3A). The particulate and supernatant fractions from soy milks containing group I also had larger amounts of glycinin than those without group I (Figure 3B−D). The particulate fractions (Figure

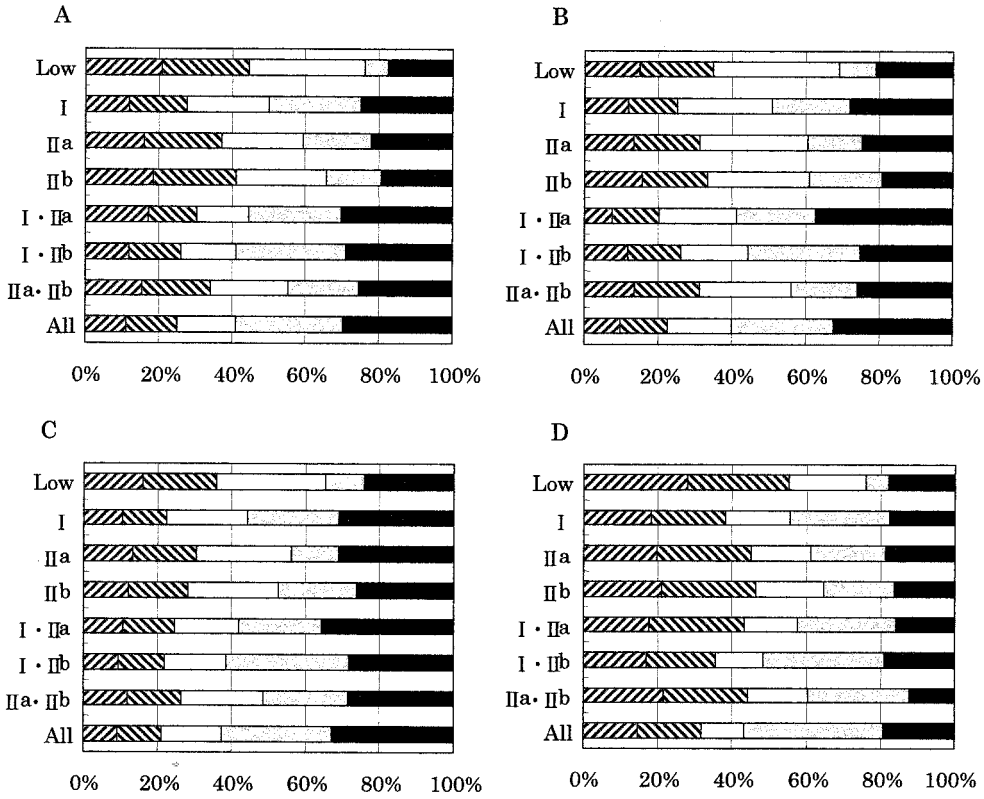

Figure 3. Subunit compositions of whole protein (A), large protein particle (B), medium protein particle (C), and supernatant protein (D) in soy milks prepared from eight soybean lines of different subunits of glycinin. These values were determined by densitometric analysis of SDS−PAGE bands. The soy milks containing 11.5% solids were prepared from eight lines having a low content of glycinin (Low), group I (I), group IIa (IIa), group IIb (IIb), group I and IIa (I·IIa), group I and IIb (I·IIb), group IIa and IIb (IIa·IIb), and group I, IIa, and IIb (All). Subunit: α′ (hatched bars), α (back-hatched bars), and β (white bars) subunits of β-conglycinin and acidic (gray bars) and basic (black bars) subunits of glycinin.

3B,C) of soy milks prepared from various soybeans contained larger amounts of glycinin than the supernatant (Figure 3D). The formation of particulate fractions must be promoted with glycinin content. It was reported that the interaction of the β-subunit of β-conglycinin and the basic subunit of glycinin was induced to aggregates by heating in a soybean protein (Utsumi et al., 1984; Arrese et al., 1991). The protein particle in soy milk contained a large amount of the β-subunit of β-conglycinin and the basic subunit of glycinin (Figure 3B,C). These results suggest that the particles in soy milk had a large amount of glycinin, which was formed by conjugation of the core of β and basic subunits (Ono et al., 1991).

The soy milks without group I contained larger amounts of β-conglycinin (Figure 3A). We observed that such soy milks contained fewer particles than those from soybeans normally used for tofu manufacturing (Tezuka et al., 1995). When β-conglycinin, a glycoprotein, is heated at a low ionic strength and neutral pH, it dissociates into subunits owing to its hydrophilic property (Thanh and Shibasaki, 1979). In contrast, glycinin has been reported to coagulate by heating at a 0.5 ionic strength and neutral pH (Wolf and Tamura, 1969), accompanying its hydrophobic region being exposed to the molecular surface. These results suggest that glycinin is essential for particle formation and that the particles play an important role in the formation of the core of the tofu curd.

Effect of Calcium and pH on Protein Solubility in Various Soy Milks. Protein solubilities of soy milks prepared from various soybeans were measured at various calcium concentrations, and the results are shown in Figure 4A. The protein solubility in soy milk prepared from soybean having a low content of glycinin (S-Low) decreased between 8 and 14 mM of calcium chloride. A decrease in solubility was observed between 6 and 12 mM for soy milks containing group IIa (S-IIa) and IIb (S-IIb), between 6 and 10 mM for those containing group I (S-I), I and IIb (S-I·IIb), IIa and IIb (S-IIa·IIb), and I, IIa, and IIb subunits (S-All). The protein solubility in soy milk containing group I and IIa (S-I·IIa) decreased most sharply between 6 and 9 mM calcium chloride. These results indicated that the protein solubility in soy milks containing group I decreased at lower calcium concentrations than those without group I. Therefore, the sensitivity to the calcium ion is higher. Ono et al. (1993) reported that the protein particulate fraction in soy milk was precipitated at lower calcium concentrations than other fractions. The higher calcium-sensitivity of soy milks containing group I may be dependent on the larger proportion of protein particles in these soy milks (Figure 2).

Figure 4B shows a relationship between protein solubility and the pH of soy milks with various concentrations of calcium. The protein solubility of soy milks containing group I (S-I, S-I·IIa, S-I·IIb, and S-All) decreased in a higher pH range between pH 6.3 and 6.0 than did the other soy milks. The soy milks containing group I had many protein particles and coagulated at a lower calcium concentration (Figure 4A). Therefore, the protein solubility of soy milk containing many protein particles decreased at low calcium concentrations and higher pH than did soy milks with fewer particles.

Effect of Magnesium and pH on Protein Solubility in Various Soy Milks. When tofu curd is manu-

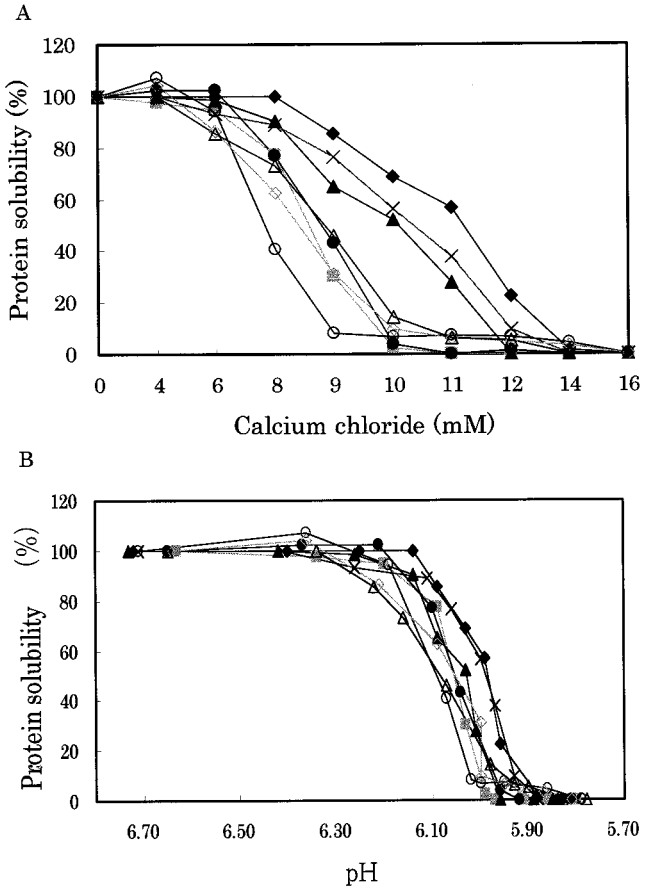

Figure 4. The changes in protein solubility (A), and the relationship between protein solubility and pH (B) in soy milk with the addition of calcium chloride. The soy milks containing 11.5% solids were prepared from eight soybean lines having a low content of glycinin (◆), group I (■), group IIa (▲), group IIb (×), group I and IIa (○), group I and IIb (●), group IIa and IIb (◇), and group I, IIa, and IIb (△).

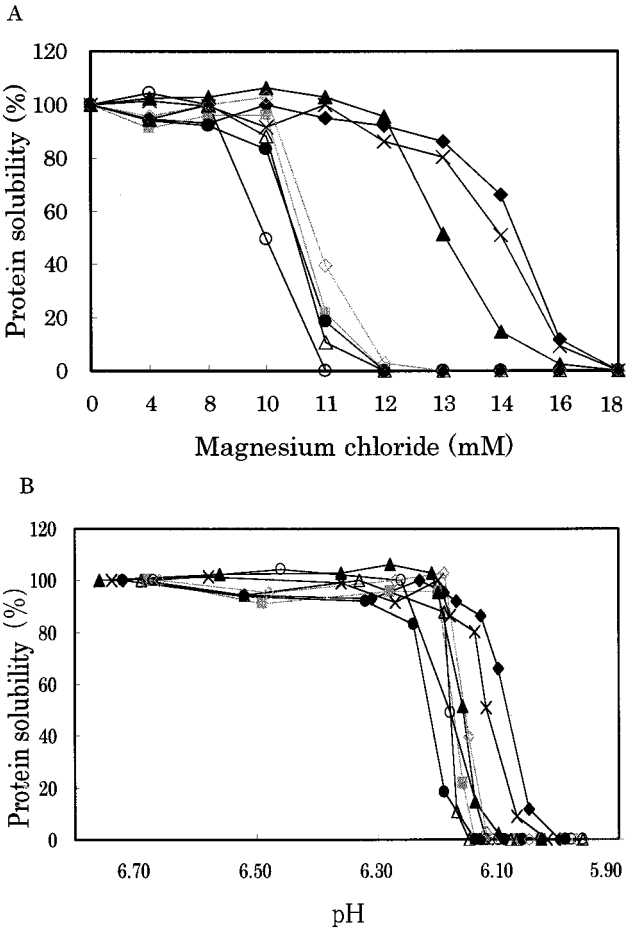

Figure 5. Changes in protein solubility (A) and the relationship between protein solubility and pH (B) in soy milk with the addition of magnesium chloride. The soy milks containing 11.5% solids were prepared from eight soybean lines having a low content of glycinin (◆), group I (■), group IIa (▲), group IIb (×), group I and IIa (○), group I and IIb (●), group IIa and IIb (◇), and group I, IIa, and IIb (△).

factured, magnesium chloride is often used instead of calcium salts. Therefore, the protein solubility of soy milk was measured with the addition of magnesium chloride. The result is shown in Figure 5A. The decease in protein solubility caused by magnesium chloride was observed at higher concentrations than that caused by calcium chloride (Figure 4A). The protein solubility in S-Low and S-IIb decreased between 14 and 16 mM magnesium chloride. The decease of protein solubility in S-IIa was observed between 12 and 14 mM and that in S-I, S-I·IIb, S-IIa·IIb, and S-All was observed between 10 and 12 mM. The protein solubility in S-I·IIa decreased at the lowest magnesium range between 8 and 11 mM. These results indicate that the protein solubility in soy milks containing group I decreased at a lower magnesium concentration than the others as well as that in the addition of calcium chloride. The soy milks containing the largest amount of group I had more protein particles and showedgreater sensitivity against calcium and magnesium ions.

Figure 5B shows a relationship between protein solubility and pH in soy milks at various magnesium concentrations. The protein solubility of S-Low and S-IIb decreased between pH 6.2 and 6.0, and that of S-I, S-IIa, S-I·IIa, S-I·IIb, S-IIa·IIb, and S-All decreased between pH 6.2 and 6.1. Ono et al. (1993) reported that the coagulation of soy milk by lowering pH with HCl occurred at pH 5.8, and that when calcium or magnesium ions were added to soy milk, coagulation started at a higher pH than that without these ions. Furthermore, coagulation accelerated by decreasing the pH. Therefore, the formation of tofu curd would be started by binding calcium or magnesium ions with protein particles, and the proteins coagulated with the lowering of the pH. The pH for protein coagulation in soy milk by magnesium chloride was 0.1 pH higher than that by calcium chloride (Figure 4B). This coagulation occurred in a narrower pH range than that in the addition of calcium chloride. These results suggest that the calcium ion is stronger and more sensitive to combine with proteins than the magnesium ion.

Distribution of the Protein Particles from Each Group. The soy milks having group I contained larger amounts of glycinin than those without group I (Figure 3A). The soy milks containing larger amounts of glycinin had more protein particles than others. Therefore, it is not clear whether the amount of protein particles in soy milk depends on the content of group I or glycinin. To elucidate this point, each group−protein was purified and added into Low soy milk to make I, IIa, or IIb group−protein soy milk having identical levels of glycinin content. The distribution of protein particles in these soy milks was measured and is shown in Figure 6. The proportions of protein particles in group−protein soy milks were higher in all cases than the respective raw group−protein soy milks. The order of particulate

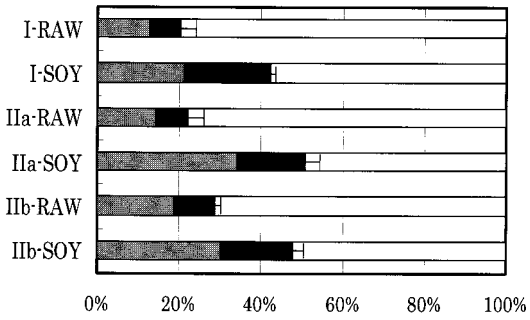

Figure 6. Distribution of the protein particles in raw group−protein soy milks (RAW) and group−protein soy milks (SOY) by differential centrifugation. The particles of more than 100 nm, 40−100 nm, and less than 40 nm in diameter were large protein particle (gray bars), medium protein particle (black bars), and supernatant protein (white bars), respectively. Values are means ± standard deviation (sum of large and medium protein particles), $n = 5$ per group. Each group protein (I, IIa, or IIb) was purified and added into Low soy milk to make I, IIa, or IIb group−protein soy milks with identical levels of glycinin content, respectively.

content was IIa, IIb, and I group−protein soy milks. The IIa group has the highest ability to form particles among these glycinin groups. Therefore, the higher proportion of protein particles in soy milks containing group I (already shown in Figure 2) is due to a higher proportion of glycinin content.

Breaking Stress of Tofu Curds from Various Soy Milks Adjusted to the Same Glycinin Content. The soy milks containing group I had more glycinin and protein particles than those without group I (Figures 2 and 3A). The breaking stress values of the tofu curds containing group I were higher than those from tofu curds without group I (Figure 1). It is known that the hardness of tofu curd is influenced by the ratio of glycinin to β-conglycinin (Saio et al., 1969). The glycinin content in various soybeans having a different glycinin subunit was not the same (Figure 3). To elucidate the influence of each group on the breaking stress, the glycinin content in soy milk was adjusted to an identical level by using a soybean mixture according to the glycinin content calculated from Figure 3A. Group I tofu curd containing 13.0% solids was prepared from a mixture of 51.5 g of soybeans having group I only and 78.5 g of soybeans having a low content of glycinin. Group IIa tofu curd containing 13.0% solids was prepared from a mixture of 80.9 g of soybeans having group IIa only and 49.1 g of soybeans having a low content of glycinin. Group IIb tofu curd containing 13.0% solids was prepared from soybeans having group IIb only. The breaking stress value of tofu curd prepared from these soy milks was measured and is shown in Figure 7. The breaking stress values of group I, IIa, and IIb tofu were 4970, 7020, and 6550 Pa, respectively. The order of breaking stress values of tofu hardness was IIa, IIb, and I. The order of particulate content in group−protein soy milks was also IIa, IIb, and I, as already shown in Figure 6. These results indicate that the hardness of tofu curd is influenced by the amount of protein particles in soy milk.

CONCLUSION

The breaking stress value of tofu curds prepared from soybeans having group I of the glycinin subunit was twice that of those without group I. The soy milks containing the largest amount of group I had the largest

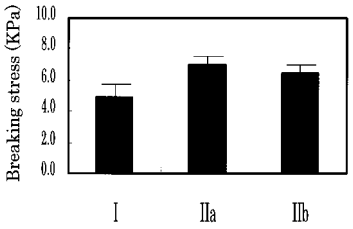

Figure 7. Breaking stress of tofu curds prepared from soy milks with identical levels of glycinin content. Values are means ± standard deviation, $n = 7$ per group. Tofu curd containing 13.0% solids was prepared by adding 2.1 mL of magnesium chloride solution (50 g of $MgCl_2 \cdot 6H_2O$ + 5.0 g of NaCl/200 mL of H_2O) from the mixture of soybeans having group I (Group I), IIa (Group IIa), and IIb (Group IIb) and soybean having a low content of glycinin.

numbers of protein particles and showed greater sensitivity to calcium and magnesium ions. The soy milks containing group I had larger amounts of glycinin and protein particles than those without group I. To elucidate the influence of each group on the breaking stress, the glycinin content and solid concentration in soy milks having each group were adjusted to an identical level. The order of hardness in breaking stress values of these tofu curds was IIa, IIb, and I. The order of particulate content in these soy milks was also IIa, IIb, and I. Therefore, the results suggest that the breaking stress value of the tofu curds is dependent upon the number of protein particles in the soy milk and that the number of the particle is determined by the proportion and structure of glycinin.

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