In: Advances in Cereal and Pseudocereal ... ISBN: 978-1-62618-347-6
Editors: N. Morita, P.V. Hung et al.

Chapter 5

GERMINATED BUCKWHEAT FOR FUNCTIONAL FOODS

Naofumi Morita[1], Kazuyoshi Miyake[2], Tomoko Maeda[3] and Pham Van Hung[4]

[1] Department of Food Packaging Technology, Toyo College of Food Technology, Mimami-Hanayashiki, Kawanishi, Hyogo, Japan
[2]Miyake Flour Milling Co., Ltd., Momodani, Ikuno-ku, Osaka, Japan
[3]Department of Science, Technology, and Human Life, Graduate School of Education, Hyogo University of Teacher Education, Shimokume, Kato, Hyogo, Japan
[4]School of Biotechnology, International University, National University in HoChiMinh City, HoChiMinh City, Vietnam

ABSTRACT

Buckwheat grains were gradually milled from the inner to the outer layers. The characteristics of 5 types of buckwheat flour obtained from milling [(special flour (FS), 1^{st}-3^{rd} grade flours (F1-F3) and seed coat flour (FBB)] were determined**.** The protein and ash content of all buckwheat fractions increased as milling progressed from the inner fraction to the outer fraction. The F3 fraction, which includes bran and the germ, was the richest in nutritional value. During germination at 25°C, both gamma-amino butyric acid (GABA) levels and the enzymatic activity of α-amylase and protease showed a greater increase than during wheat germination**.** To decrease the allergenicity of buckwheat grains,

buckwheat germination was performed and the characteristics of the grains were assessed. Germinated buckwheat grains were fermented to generate the processed foods, such as soba *natto* and soba *miso* paste. For soba *miso* paste, the paste became dark with red and yellow tints due to the Maillard browning reaction, the pH value decreased gradually (pH 6.25 to 5.6) after storage for 60 days, and the total titratable acidity (TTA) value increased (3.8 to 9.5) according to the length of fermentation. During fermentation, the amounts of GABA produced in soba *natto* and *miso* paste 48 hr and 60 days of fermentation, respectively, were 3.3 and 1.7 times higher than at the start of fermentation. SDS-PAGE analysis of albumin and globulin from soba *natto* during fermentation indicated that high molecular weight proteins (45-97 kDa) were less abundant after 48 hr of fermentation, and low molecular weight (LMW) proteins were generated. Allergenic proteins were identified with IgE immunoblotting, and the majority of the allergenic 12 and 22 kDa proteins from albumin and globulin were degraded to LMW proteins. The HMW albumin and globulin proteins from soba *miso* paste decomposed after 60 days of fermentation, and the ability of the proteins to bind to IgE also decreased. For commercial-scale germination of buckwheat grains, heating and humidifying of moist air methods was applied rather than soaking directly in water.

As a result, germinating buckwheat might improve the properties of the grain, leading to a higher nutritional value, improved flavor, and decreased allergenicity.

INTRODUCTION

The word “buckwheat” contains “wheat,” but buckwheat is not a cereal. The plant is classified as a pseudocereal and a dicotyledonous plant. The production of buckwheat grain has many drawbacks compared to rice or wheat due to gramineous plants. Therefore, cultivation today is limited to highland or alpine valleys where rice and wheat cannot be grown. However, among grain crops, buckwheat is gaining popularity because it contains bioactive components with beneficial health properties. Additionally, buckwheat proteins are composed of well-balanced amino acids with a high amino acid score and have anti-cholesterolemic properties (Tomotake et al., 2000). Buckwheat starches are less easily digestible, affording a low glycemic index (Skrabanja et al., 2001). Furthermore, many of the health benefits of the buckwheat grain are correlated to the presence of gamma-amino butyric acid (GABA), the flavonoids rutin and quercetin, and other phenolics. Rutin has been reported to alleviate many modern health disorders caused by the

nutritional habits currently prevailing in western countries. The beneficial properties of rutin include antimutagenic activity (Undeger, 2004), protective effects against the development of diabetes (Srinivasan et al., 2005), antioxidative properties (Oomah & Mazza, 1996, Hung & Morita, 2008), a well-balanced amino acid composition with a high biological value (Pomeranz, 1983), and a decrease in plasma cholesterol levels (Kayashita et al., 1997). However, another property of buckwheat critical to human health is the presence of proteins that cause a hypersensitive allergic reaction.

Since the development of the modern milling apparatus, refined buckwheat flour contains little of the fiber, minerals and phenolic compounds found in the bran and germ. Therefore, we are obliged to eat buckwheat flour that is less nutritious than it could be. Recently, a gradual reduction milling system was developed to improve nutritional content and to remove allergenic proteins from flour. We also germinated buckwheat grains to decrease the allergenic protein content and to increase the nutritional properties of buckwheat. The purpose of the present study was to determine the characteristics of various graded fractions obtained by milling the germinated buckwheat cultivar Mancan and also to characterize changes in the properties of germinated buckwheat flour. These results can be applied to develop milling procedures for buckwheat grains that yield buckwheat products with low- or no-allergenicity.

GRAIN AND FLOUR

The buckwheat grain used for germination was the cultivar Mancan, which was imported from China and supplied by Miyake Flour Milling Co., Ltd. (Osaka, Japan). The buckwheat grain was gradually milled from the innermost portion of the grains; the five fractions FS, F1, F2, F3 and FBB were obtained in order from the innermost to the outermost fractions**,** as shown in Fig. 5-1.

Flour quality was tested using ICP (Inductively Coupled Plasma) analysis to assess inorganic compounds and amino acid content. Scanning electron microscopy (SEM) was used to observe the starch granules of the different fractions of flour prepared from germinated grains. Whole buckwheat grains (groats) were soaked in water for 1 hr at 25°C, then germinated for the specified time with a relative humidity of 85% at 25°C. For comparison, the wheat grain ICW was germinated under the same conditions, except the

temperature was 30°C. After germination for the specified time interval, the buckwheat grains were collected, frozen, and lyophilized.

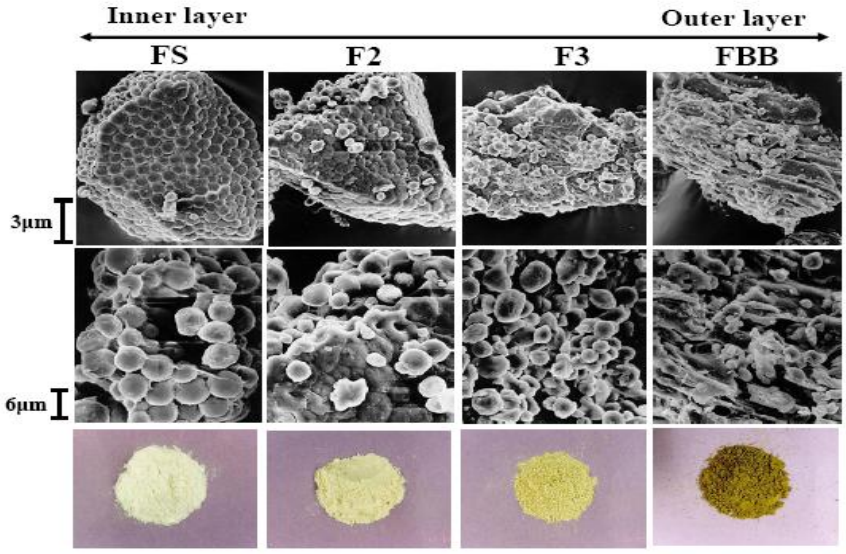

Figure 5-1. SEM images of graded buckwheat flours (adapted from Miyake et al., 2004). FS (×1000, 2500); F2 (×1000, 2500); F3 (×1000, 2500); FBB (×1000, 2500).

CHARACTERISTICS OF GERMINATED BUCKWHEAT FLOUR

Table 5-1 shows a proximate analysis of the buckwheat flour. The moisture content of buckwheat flours decreased in the order FS, F1, F2, F3 and FBB, whereas the amount of ash increased in the same order, except for the FBB fraction. The amount of protein, lipid and ash increased from the inner fraction FS to the outer fraction F3. FBB, the outermost fraction, contained the largest amount of carbohydrates.

The mineral content of the inner fractions FS, F1 and F2 contained smaller amounts of Ca, whereas the outer F3 and FBB fractions contained larger amounts (Table 5-2). All fractions contained large amounts of K, Mg and P, and the FBB fraction in particular contained the largest amount of S among all the fractions tested**.**

The amount of phytic acid (IP6) in various germinated flour samples was measured using the spectrophotometric method at 500nm (Latta and Eskin, 1980). During germination for 24 hr, the amount of IP6 detected from

buckwheat did not change appreciably (0.44-0.47%), whereas the amount measured in wheat increased significantly (0.07-0.35%), reaching approximately 5 times the starting amount (data not shown).

Table 5-1. Proximate analyses of buckwheat fractions (adapted from Miyake et al**., 2004)**

Sample	Moisture	Ash	Protein	Lipid	Carbohydrate
			(%)*		
FS	14.4	0.2	3.8	2.6	79.0
F1	14.3	0.5	5.5	3.0	76.7
F2	12.4	3.1	23.7	5.9	55.0
F3	10.9	5.2	38.1	8.8	37.0
FBB	7.2	2.6	4.0	2.8	83.4

FS, F1, F2, F3 and FBB were gradually milled from the innermost to the outermost fraction of a whole buckwheat grain. FS, special flour; F2-F3, 2nd~3rd grade flours; FBB, seed coat flour.

* Values were calculated on a dry flour basis.

Table 5-2. Mineral components of graded buckwheat flours (adapted from Miyake et al**., 2004) (mg/100g of dry flour)**

Element	FS	F1	F2	F3	FBB
B	0.27	0.18	0.79	1.43	0.54
Ca	0.001	0.001	0.002	43.4	117.6
Cu	0.13	0.14	0.32	0.73	0.33
Fe	0.30	0.48	2.73	5.03	1.27
K	140.7	190.4	1085.5	2208.9	905.6
Mg	22.8	34.8	228.3	432.2	85.4
P	52.6	92.3	597.0	1129.0	66.7
Pd	0.04	0.04	0.04	0.04	0.05
Rb	0.95	1.08	3.97	7.60	1.68
S	1.08	0.89	0.35	0.59	2.40
Se	0.06	0.11	0.10	0.01	0.08
Sn	0.00	0.01	0.00	0.02	0.00
Zn	0.27	0.42	2.34	4.41	0.52

Abbreviations are the same as in Table 5-1.

AMINO ACID COMPOSITION OF GRADED FLOURS AND GERMINATED GRAINS

From the amino acid composition of the various buckwheat fractions shown in Table 5-3, it is apparent that the F3 fraction containing the germ and seed coat had larger quantities of amino acids compared to the other fractions. In addition, F3 clearly contained a larger amount of GABA than the other samples. Although the FBB fraction is similar to the F3 fraction in relative amino acid composition, the FBB fraction contained strikingly decreased quantities of amino acids compared to F3. However, the amount of GABA in FBB was higher than the FS, F1 and F2 fractions.

Table 5-3. Amino acid components in graded buckwheat flours* (adapted from Miyake et al**., 2004) (mg/100g of dry flour)**

Amino acid	FS	F1	F2	F3	FBB
Asp	61	467	1603	2614	154
Thr	239	218	574	1052	150
Ser	302	312	991	1624	215
Asn	13	21	581	0	0
Glu	1535	1246	4964	8471	1461
Pro	195	99	1029	2133	189
Gly	646	567	1179	2027	333
Ala	505	300	900	1349	155
Val	301	33	887	1412	102
Cys	61	73	604	1016	52
Met	25	168	445	668	70
I-leu	250	162	655	1199	1.9
Leu	454	374	1275	2181	196
Tyr	188	177	473	873	13
Phe	311	215	747	1211	90
GABA	9	3	87	310	89
Orn	128	20	66	15	1
Lys	500	370	1039	1800	224
His	184	767	362	547	28
Arg	641	419	2119	3870	91
Total amounts	7107	6531	21774	34954	4354

Abbreviations are the same as in Table 5-1.

*All data were collected by hydrolysis with hydrochloric acid.

Germination of buckwheat grain generated higher levels of amino acids**,** especially GABA, which increased 2.5 times after incubation for 24 hr compared to without germination, as shown in Table 5-4. By comparison, GABA increased 1.5 times under the same conditions in germinating wheat grains.

Table 5-4. Changes to free amino acid content of buckwheat and wheat grains during germination (adapted from Miyake et al**., 2004)**

Amino acid	Buckwheat (mg/100g of dry flour)				Wheat (mg/100g of dry flour)			
	Control	G-8	G-16	G-24	Control	G-8	G-16	G-24
THR	13.4	18.2	21.3	29.5	4.1	5.6	12.8	12.6
VAL	5.5	4.7	6.5	10.4	5.7	6.8	15.2	16.1
MET	8.8	9.9	10.3	13.2	2.1	5.9	5.1	5.3
LEU	11.7	13.2	13.0	16.9	3.8	7.2	16.8	15.9
TYR	8.7	11.2	9.9	10.7	4.0	3.6	11.5	12.5
PHE	10.2	11.9	13.2	16.8	3.3	4.8	11.6	12.0
HIS	5.6	10.6	13.7	23.2	2.7	4.0	9.2	8.5
LYS	9.6	10.4	13.2	19.6	9.2	10.4	12.8	17.6
GLU	72.4	75.4	105.6	133.4	36.7	59.6	94.1	82.6
GLN	16.5	27.3	44.6	88.4	13.7	5.0	34.5	34.6
GABA	12.4	11.6	22.5	28.7	5.2	3.6	6.9	7.9
Total	174.8	204.4	273.8	390.8	90.5	116.5	230.5	225.6

In terms of the α-amylase and protease activity during germination, buckwheat is more active than wheat grain. α-Amylase from buckwheat showed a peak in activity after incubation for 16 hr, and protease activity increased steadily for the length of incubation, as reported by Kikunaga and Takahashi (1992).

As described in the results above, each graded flour fraction possessed different flour qualities and different functional and nutritional properties. It is well known that the amino acid score of buckwheat is much higher than rice or wheat because buckwheat contains a large amount of lysine and tryptophan. Therefore, germination experiments were performed with the expectation that the level of allergenic substances in buckwheat would decrease during the germination process. In fact, *Rhizopus oligosporus* has been known to increase the functional properties of tempeh, which is made from buckwheat or soybean, and was very effective at reducing the allergenic protein composition

of buckwheat, as reported by Handoyo et al. (2006) and Kobayashi et al. (2004). For practical food applications, buckwheat grains were used for the preparation of soba *natto* and *miso* paste, both typical and traditional foods in Japan. *Natto* is highly regarded worldwide as a healthy food.

PREPARATION OF SOBA *NATTO* AND *MISO* PASTE USING GERMINATED BUCKWHEAT

Germinated and autoclaved buckwheat was used for the preparation of soba *natto* and *miso* paste. Soba *natto* was prepared by inoculating the buckwheat with *Bacillus natto* spores. Fermentation occurred for 0, 12, 24, 36 or 48 hr at 40^{o}C using a commercial "Domestic *natto* maker" apparatus according to the manufacturer's instructions (Tokyo Unicom Co. Ltd., Tokyo, Japan). Soba *miso* paste was prepared using pre-germinated buckwheat (450g), malted rice (300g), salt (110 g) and water (50g), then stored in an incubator at 30^{o}C for 0, 15, 30, 45 or 60 days.

CHARACTERISTICS OF SOBA *NATTO* AND *MISO* PASTE MADE FROM GERMINATED BUCKWHEAT

During soba *natto* fermentation, the protein content noticeably decreased; therefore, the characteristics of the proteins in soba *natto*, including molecular weight and stability, were changed during fermentation.

For soba *miso* paste, the appearance of the paste was considerably changed to a darker color due to the Maillard browning reaction. After storage for 60 days, the soba *miso* paste was dark with a red and yellow tint. The pH value decreased gradually (pH 6.25 to 5.6), and the total titratable acidity (TTA) value increased (3.8 to 9.5) according to the length of fermentation.

AMINO ACID CHANGES IN SOBA *NATTO* AND *MISO* PASTE

During fermentation, the amount of GABA produced in soba *natto* and *miso* paste clearly increased, particularly the amounts of GABA in soba *natto* and *miso* paste after 48 hr and 60 days of fermentation, which were 3.3 and 1.7

times higher than at the start of fermentation, respectively (Tables 5-5 and 5-6).

Table 5-5. Amino acid composition of soba *natto* during fermentation (adapted from Miyake et al**., 2006) (mg/100mL)**

Amino acid	Fermentation time (hr)				
	0	12	24	36	48
Asp	3.4	1.3	4.1	7.4	6.7
Thr	7.2	1.9	2.2	1.1	3.3
Ser	7.8	4.6	2.5	1.0	1.8
Asn	3.3	1.3	2.6	2.2	0.8
Glu	47.8	17.4	12.3	7.6	12.8
Pro	4.7	1.8	5.7	3.6	7.8
Gly	3.8	2.1	0.7	0.1	0.6
Ala	9.9	10.1	4.5	1.6	3.9
Val	4.3	2.9	3.4	2.8	3.6
Cys	8.7	6.0	12.0	6.7	12.2
Met	1.0	0.7	1.3	1.4	5.1
I-leu	42.0	34.2	32.8	17.4	19.9
Leu	16.4	11.5	22.8	3.4	9.9
Tyr	3.5	3.0	4.0	4.4	5.4
Phe	3.8	2.9	4.0	4.9	5.4
GABA	2.6	7.6	6.9	6.5	8.6
Etha	3.6	4.1	12.1	5.8	18.3
Ammo	5.9	4.0	11.4	9.3	15.7
Orn	0.3	1.8	8.7	3.3	10.1
Lys	5.8	3.5	5.6	5.3	5.5
His	12.8	9.5	11.5	10.2	13.8
Arg	31.6	17.2	10.4	3.6	3.5
Total	230.5	149.5	181.6	109.5	174.8

In addition, glutamic acid levels in soba *natto* decreased (0.27 times), although in soba *miso* paste these levels increased (5.7 times). Because GABA is generated from glutamic acid by glutamate decarboxylase, the decrease in glutamic acid levels in soba *natto* during fermentation might be caused by GABA formation. In the fermentation of soba *miso* paste, the amount of glutamic acid produced was higher than the amount consumed by GABA formation during fermentation. The total amount of amino acids in soba *miso* paste also increased, but in soba *natto* the amino acid levels decreased during fermentation. Therefore, in the production of soba *miso* paste, new amino acids were produced rather than consumed during fermentation; however, the

amino acids were substantially consumed during soba *natto* fermentation, resulting in lower levels when compared to the start of fermentation. This result correlated with decreasing protein levels during fermentation.

Table 5-6. Amino acid composition of soba *miso* paste during fermentation (adapted from Miyake et al., 2006) (mg/100mL)

Amino acid	Fermentation time (days)				
	0	15	30	45	60
Asp	0.6	7.0	22.3	31.0	21.9
Thr	1.2	7.9	10.0	10.8	7.8
Ser	2.1	9.1	19.8	24.1	18.4
Asn	1.0	9.1	10.1	9.6	12.0
Glu	11.7	53.5	72.4	81.8	66.8
Pro	3.3	8.3	10.3	11.1	8.7
Gly	0.3	7.4	17.0	21.6	15.9
Ala	14.6	26.8	49.3	52.1	43.6
Val	1.4	8.4	18.8	21.0	15.8
Cys	5.5	9.6	7.6	9.4	19.4
Met	0.7	3.1	1.9	2.0	3.9
I-leu	18.7	50.8	19.7	20.6	72.7
Leu	10.2	34.1	30.1	31.4	51.8
Tyr	2.3	7.5	14.9	16.1	11.3
Phe	0.5	8.4	9.5	7.2	8.7
GABA	6.5	7.9	4.9	4.2	10.9
Etha	34.9	41.3	17.2	23.9	69.1
Ammo	7.0	11.0	8.2	8.9	16.6
Orn	12.8	5.5	7.6	7.0	5.0
Lys	13.5	44.9	46.3	49.0	58.2
His	1.8	8.2	12.3	13.3	11.2
Arg	11.1	53.9	68.1	73.9	73.4
Total	161.8	423.8	478.3	530.1	623.1

DEGRADATION OF ALLERGENIC PROTEINS IN BUCKWHEAT DURING FERMENTATION

SDS-PAGE analysis of albumin and globulin proteins from soba *natto* during the fermentation time course is shown in Fig. 5-2.

The protein composition at the start of fermentation (0 hr of fermentation) shows proteins of various molecular weights, but high molecular weight (HMW) proteins were degraded during the time course of fermentation. The

protein bands corresponding to albumin (45-97 kDa) at 48 hr decreased compared to 0 hr. For globulin, a protein band of approximately 45 kDa was faint, but low molecular weight (LMW) proteins of approximately 22 kDa were produced after fermentation for 48 hr, compared to the sample at the start of fermentation (0 hr). These results suggest that *B. natto* rapidly degraded the HMW proteins and utilized them for the production of amino acids or LMW peptides for microbial growth.

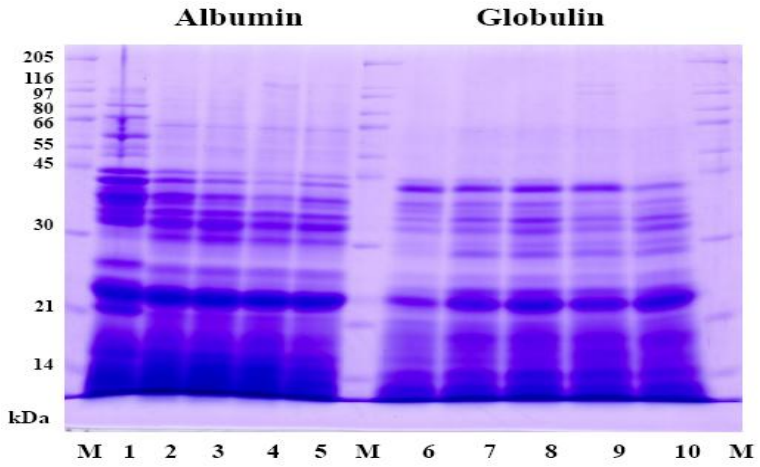

Figure 5-2. SDS-PAGE of albumin and globulin proteins extracted from soba *natto* samples after fermentation (adapted from Miyake et al., 2006). M is the molecular weight marker. 1, albumin (0); 2, albumin (12); 3, albumin (24); 4, albumin (36); 5, albumin (48); 6, globulin (0); 7, globulin (12); 8, globulin (24); 9, globulin (36); 10, globulin (48). Parenthetical values correspond to the fermentation time (hr).

The quantity of allergenic proteins in buckwheat samples was examined with immunoblotting analysis using an IgE that specifically binds to the proteins related to buckwheat allergy, as shown in Fig. 5-3. IgE bound most strongly to the 12 and 22 kDa albumin and globulin proteins in the control soba *natto* without fermentation (0 hr), and binding decreased after fermentation for 36 hr. According to a report by Yoshioka et al. (2004a, 2004b), the 22 kDa protein is a major allergen in Mancan buckwheat. Western blot analysis showed that the allergenic proteins were present in the control without fermentation (lanes 1 and 4 in Fig. 5-3) and were clearly albumin protein. However, these bands disappeared over the time course of fermentation (lanes 3 and 6 in Fig. 5-3). Therefore, in soba *natto* after 36 hr of

fermentation, the allergenic albumin proteins were mostly degraded, and the allergenic globulin proteins were completely degraded to LMW proteins (amino acids or small peptides) (lanes 3 and 6 in Fig. 5-3).

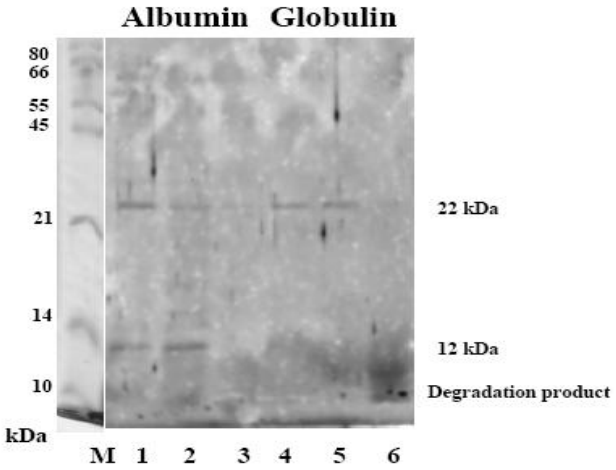

Figure 5-3. IgE immunoblotting of albumin and globulin proteins extracted from soba *natto* samples after fermentation (adapted from Miyake et al., 2006). M is the molecular weight marker. 1, albumin (0); 2, albumin (24); 3, albumin (36); 4, globulin (0); 5, globulin (24); 6, globulin (36). Parenthetical values correspond to the fermentation time (hr).

SDS-PAGE analysis of albumin, globulin, glutelin and prolamin in soba *miso* paste after fermentation for 0, 30 and 60 days are shown in Fig. 5-4.

The HMW protein bands from albumin, globulin and glutelin were weak and disappeared after 60 days of fermentation, although prolamin was an exception. Immunoblotting analysis of albumin and globulin after fermentation for 60 days demonstrated that the proteins of approximately 15 and 22 kDa disappeared after fermentation for 60 days (lanes 3 and 6 in Fig. 5-5).

From these results, it appears that the HMW albumin and globulin proteins from soba *natto* and *miso* paste were decomposed, and consequently, LMW proteins were produced during fermentation. In addition, the binding of IgE to these proteins, assessed by immunoblotting, became weaker over the long fermentation period, although the total amount of LMW proteins increased. Therefore, germinating buckwheat was effective, suitable and useful for the production of soba *natto* and *miso* pastes with low- or no-allergenicity. Furthermore, the germinated buckwheat can be used for the

production of germinated-buckwheat products, such as soba *natto* and *miso* paste, which are a new type of fermented food distinct from ordinary *natto* and *miso* paste, which are made from soybeans.

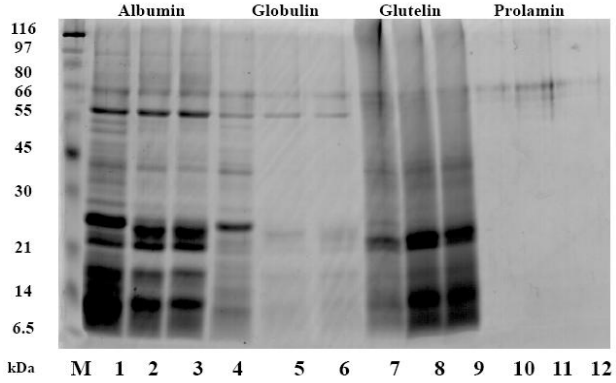

Figure 5-4. SDS-PAGE of proteins extracted from soba *miso* paste after fermentation (adapted from Miyake et al., 2006). M is the molecular weight marker. 1, albumin (0); 2, albumin (30); 3, albumin (60); 4, globulin (0); 5, globulin (30); 6, globulin (60); 7, glutelin (0); 8, glutelin (30); 9, glutelin (60); 10, prolamin (0); 11, prolamin (30); 12, prolamin (60). Parenthetical values correspond to the fermentation time (days).

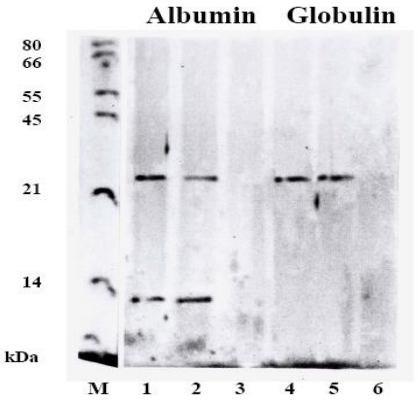

Figure 5-5. Immunoblotting analysis of albumin and globulin proteins extracted from soba *miso* paste after fermentation (adapted from Miyake et al., 2006). M is the molecular weight marker. 1, albumin (0); 2, albumin (15); 3, albumin (60); 4, globulin (0); 5, globulin (15); 6, globulin (60). Parenthetical values correspond to the fermentation time (days).

IMPROVED GERMINATION METHODS FOR CEREAL GRAINS

Recently, the preparation of germinated cereal grains avoided direct soaking of the grains in water because beneficial low molecular weight materials formed during germination were released into the solution. Therefore, minute amounts of water were used for germination. This method for GABA production was proposed as a fine moistening method by Satake Co. Ltd as the "Satake Co. method." (Satake et al., 2003; Satake, 2007). These methods were originally used for the germination of brown rice. The fine moistening method for brown rice hydrates the grains at 30°C and 19%-23% moisture (increased speed: 0.5%/hr for a grain moisture range below 17%, 0.5% to 1.2%/hr for a grain moisture over 17%, tempered for 12 hr) for 1 to 2 days. Using the "Satake Co. method," the grains contain 18mg GABA/100g dry basis, whereas after the water immersion method for 24 hr at 20~25°C, the grains contain 10~15mg GABA.

This method was further improved using a higher temperature (60-75°C), a shorter time (approximately 5 hr) with moist air (95% relative humidity) and termed the "heating and humidifying of moist air method" (Naka et al., 2011). The amount of GABA formed was approximately 3-fold (39.3mg) higher than control rie-Tartary buckwheat (13mg/100g dry basis).

Both the fine moistening method and the heating and humidifying of moist air method are convenient for the downstream milling process because extensive drying not required after germination. Additionally, these methods produce few heavy grain cracks, which is relevant to rice grain polishing.

CONCLUSION

Buckwheat has been used worldwide in processed foods, such as noodles, crepes, pasta, and some cakes for many years, and these products are usually made by mixing buckwheat flour with water and salt, followed by baking. In this study, we show that whole buckwheat grains can be used for processed foods without removing the germ, bran and seed coat (husk) after germination of the whole grain. Therefore, it is theoretically possible to utilize 100% of the grain, abrogating the need to discard byproducts such as the germ, bran and husk generated from the conventional system of buckwheat milling. In addition, germination was found to increase the value of buckwheat as a

foodstuff because the amounts of GABA, minerals, amino acids, and other beneficial nutrients increased. This implies that health-conscious people would prefer to eat food made from germinated grains due to their increased nutrient content, such as buckwheat *natto* and *miso* paste. *Natto* and *miso* paste are traditional foods in Japan, and *natto* and *miso* paste have also become international products, preferred and consumed by many foreign people as well. Furthermore, the results of this study indicate that the germination treatment decreased the allergenic protein content in the buckwheat grains, allowing the production of foods with low- or no-allergenicity. Consequently, germinating buckwheat improves its properties, leading to a higher nutritional value, better taste and decreased allergenicity. In the future, we expect that germinated-buckwheat *natto* and *miso* paste will be consumed daily as healthy food choices.

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