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REVIEW

The Effect to the Protein Concentration and Flour Quality of Nitrogen Fertilization at 10 Days after Heading in Wheat

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Abstract

To increase the protein concentration of wheat grain, we applied nitrogen fertilizer at 10 days after heading and examined its effect on agronomic characteristics and flour quality. This top dressing did not affect the agronomic characteristics except for delaying of the ripening stage, but increased the 1,000 kernel weight and test weight. Protein concentration increased linearly as the amount of applied fertilizer increased, at a rate of about 0.5% per 1 gm^{-2} nitrogen top-dressed. Gluten concentration increased as the protein concentration increased, irrespective of cultivar, breeding line and year. There was a significant negative correlation between protein concentration and flour color. As the protein concentration increased, flour color became gray. Nitrogen fertilizing at 10 days after heading effectively increased the protein concentration in wheat.

Discipline: Crop production
Additional key words: gluten, top dressing

Introduction

Yearly consumption of wheat in Japan is approximately 32 kg per capita, which mostly relies on the imports from Australia, Canada and USA. As domestic wheat cultivation is important from the viewpoint of food self-sufficiency, it has been encouraged by the Japanese government together with soybean and feed crop cultivation.

Although a grain protein concentration of 10–11% (flour protein concentration of 8.5–9.5%) is required for Japanese noodle wheat, wheat grains produced in paddy fields of the western region of Japan have a protein concentration of as low as 7–8%. The immediate improvement of nitrogen concentration is, therefore, required urgently, and higher protein concentration is also requested for bread wheat.

Grain protein concentration can be improved by, for example, the use of cultivars genetically containing high protein and the improvement of nitrogen top dressing methods. Lack of high protein cultivars in the region, however, seems to establish the fertilization method as the only way to improve grain protein concentration at the moment.

Flour quality might deteriorate if the timing of top dressing is mistaken, because top dressing at inappropriate timings not only elongates culm length and induces lodging but also causes the late-emerging head to bear greenish immature grains[3]. The experiment conducted so far showed that for late top dressing before the booting stage, a grain protein concentration increase of only 0.1%–0.2% had been achieved per 1 kg/10 a of top dressing nitrogen[4,9]. It is therefore difficult to say that top dressing before heading could effectively improve grain protein concentration.

The aim of this study is to establish a method of nitrogen top dressing on wheat at 10 days after heading that could effectively improve grain protein concentration without deterioration in quality.

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Materials and methods

Three wheat cultivars: Shirasagikomugi, Chikugoizumi, and Norin 61, and two breeding lines, Chugoku 146 and Chugoku 147 (during 1997 to 1998) were grown in the three seasons from 1996 to 1998 at Fukuyama in Hiroshima Prefecture, Japan. Experimental plots were 7.4 m long and 14 cm wide with 0.7 m spacing. Seven hundred and twenty-five seeds were planted per hill. Table 1 presents the time and amount of nitrogen fertilized. All plots were fertilized at the same rate of nitrogen except at 10 days after heading. The nitrogen top dressing treatments of 0, 4 and 8 g m^{-2} were applied at 10 days after heading. Main plots were top dressing at 10 days after heading and subplots were cultivars and breeding lines. The experimental design was a split-plot with no replications except in 1998. Cultivars and years were used as replications and analysis of variance was performed.

Heading date and ripening date were recorded. Harvest area was 4.9 m^2. Culm length and ear length were measured. Ear number per m^2 was determined. Grain yield was measured after threshing, cleaning and drying. The 1,000 grain-weight, test weight and grain protein concentration were measured. Wheat was milled through a Buhler Test Mill after conditioning for 24 h to 14.5% (w/w) grain moisture content. Flour yield and milling characteristics were measured. Starch viscosity was measured using the Brabender Amylograph. Grain and flour protein concentrations were measured by combustion method. Gluten concentration was measured using the Glutomatic System. Flour color was measured using a photoelectric reflectance colorimeter. Ash content and Farinograph dough properties were obtained using a method similar to the AACC Standard Method.

Results

Table 2 shows the results of agronomic characteristics. Ripening date was delayed by 1 to 2 days compared with the standard by nitrogen top dressing at 10 days after heading except for Chikugoizumi. Culm length and ear number were not affected. The 1,000 kernel weight and test weight increased with an increase in the amount of nitrogen top-dressed at 10 days after heading. There was a significant difference between grain yield and amount of nitrogen top-dressed in Chugoku 146 and Chugoku 147. There were significant differences in all characteristics among cultivars and breeding lines.

Fig. 1 shows the relationship of the amount of nitro-

Table 1. Amount of nitrogen (gm^{-2}) in each plot

	Basal	January	10 days after heading	Name of plot
Standard fertilization plot	6[a)]	3	0	0
10 days after heading plot				
Low N plot	6	3	4	4
High N plot	6	3	8	8

a): Nitrogen gm^{-2}.

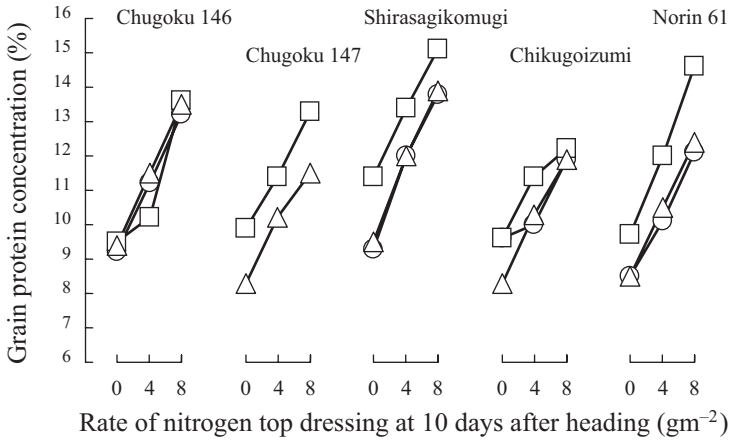

Fig. 1. The relationship between the amount of nitrogen top dressing at 10 days after heading and grain protein concentration

—○—: 1996, —□—: 1997, —△—: 1998.

Table 2. Agronomic characteristics of each plot by top dressing at 10 days after heading

Cultivar or breeding line	Name of plot	Heading date	Ripening date	Culm length (cm)	Ear no. ($/m^{-2}$)	Grain yield (gm^{-2})	1,000 Kernel weight (g)	Test weight (gL^{-1})	Grain protein concentration (%)
Chugoku 146	0	13 April	30 May	87	375	416	33.9	777	9.4
	4	13 April	31 May	86	430	436	35.7	785	11.0
	8	13 April	1 June	87	421	522	36.3	788	13.4
	ANOVA[a)]	ns	*	ns	ns	*	**	**	**
Chugoku 147	0	11 April	28 May	88	427	450	34.3	767	9.1
	4	11 April	29 May	83	432	591	37.2	788	10.8
	8	10 April	30 May	88	455	479	38.1	789	12.4
	ANOVA	ns	*	ns	ns	*	**	*	**
Shirasagikomugi	0	15 April	1 June	99	407	428	36.9	772	10.1
	4	15 April	1 June	99	372	397	39.0	786	12.5
	8	15 April	2 June	99	414	474	39.6	791	14.3
	ANOVA	ns	ns	ns	ns	ns	*	*	**
Chikugoizumi	0	11 April	31 May	92	436	481	39.7	783	9.2
	4	10 April	31 May	94	475	496	41.5	796	10.6
	8	10 April	31 May	91	473	556	42.2	798	12.0
	ANOVA	ns	ns	ns	ns	ns	*	*	**
Norin 61	0	16 April	3 June	98	428	475	38.8	783	8.9
	4	15 April	4 June	97	452	489	40.6	802	10.9
	8	15 April	5 June	95	493	439	40.8	807	13.0
	ANOVA	ns	*	ns	ns	ns	*	**	**
ANOVA[b)]									
Cultivar (C)		**	*	**	*	**	**	**	*
Amount of nitrogen (AN)		ns	**	ns	ns	ns	**	**	**
C × AN		ns	ns	ns	ns	**	ns	**	ns

Rate of nitrogen: Amount of nitrogen top dressing at 10 days after heading.
Three years average value except for Chugoku 147, Chugoku 147 is two years (1997 and 1998) average value.
*, **: Significant at the 5 and 1 % probability levels, respectively. ns: Not significant.
a): Year was used as replications.
b): Year was used as replications, all cultivars and breeding lines.

gen top-dressed at 10 days after heading and grain protein concentration. Protein concentration of 60% extraction flour increased linearly as the amount of applied fertilizer increased for all cultivars and breeding lines throughout the three years, there was a significant difference at the 1% level. The protein concentrations of 1996 and 1998 were almost the same, but lower than that of 1997. The grain protein concentrations of Chugoku 147 and Chikugoizumi were a little lower than that of the other cultivars and breeding line.

Flour yield, milling characteristics and flour quality in 1997 were excluded from the results, because the flour mill was not tuned up well. Table 3 shows the results of milling characteristics. There was a significant difference among the amount of top dressing in the BM rate and semolina yield as nitrogen fertilizations increased. The grain became stiff as the protein concentration increased (BM rate lower), and semolina yield increased. However, there were no significant differences among the amount of top dressing in flour yield and milling score. There was hardly a significant difference recognized among cultivars and breeding lines.

Table 4 shows the results of flour quality characteristics. There was a significant difference among amount of top-dressed nitrogen in grain ash of Chugoku 146, but not in the other cultivars and breeding line. The flour ash did not change significantly. There was hardly a significant difference recognized among cultivars and breeding

Table 3. Milling characteristics of each plot by top dressing at 10 days after heading

Cultivar or breeding line	Name of plot	Flour yield (%)	Milling score	BM[b] rate (%)	Semolina yield (%)
Chugoku 146	0	73.0	84.6	64.2	52.4
	4	73.9	84.2	60.3	53.6
	8	72.6	83.3	55.3	55.3
ANOVA[a]		ns	ns	ns	ns
Chugoku 147	0	74.1	83.3	66.5	54.6
	4	74.5	83.3	55.4	59.3
	8	72.9	82.8	66.9	56.1
ANOVA		–	–	–	–
Shirasagikomugi	0	74.3	85.3	66.3	53.0
	4	72.9	83.7	64.6	53.8
	8	71.7	83.4	61.9	56.7
ANOVA		ns	ns	ns	ns
Chikugoizumi	0	72.4	84.4	73.3	50.9
	4	73.6	84.5	64.6	53.6
	8	72.4	84.5	55.9	56.7
ANOVA		ns	ns	ns	**
Norin 61	0	72.6	82.5	73.6	51.6
	4	72.9	81.8	67.4	52.8
	8	70.8	82.0	60.8	54.2
ANOVA		ns	ns	ns	ns

Average value of 1996 and 1998, Chugoku 147 is only 1998.
*,**: Significant at the 5 and 1 % probability levels, respectively. ns: Not significant.
a): Year was used as replications. b): The rate of Break flour to Milling flour.

lines. In every cultivar and breeding line, there were significant differences in grain protein concentration, flour protein concentration, and concentration of gluten. L* and b* in flour color decreased and a* decreased with increasing protein concentration. The averaged correlation coefficients between these parameters and protein concentrations of two years were as follows:

$$L^*: r = -0.71,\ a^*: r = 0.81,\ b^*: r = -0.83$$

There was no significant difference among the amount of top-dressed nitrogen in maximum viscosity (MV) of the Amylograph. The falling number of 1997 was lower than that of 1998, and Chikugoizumi was lower than that of other cultivars and breeding lines. Among the amount of top-dressed nitrogen, there were significant differences in absorption (Ab), weakness (Wk), and varolimeter value (VV) of the Farinogram in every cultivar and breeding line. Wk decreased and VV increased with increasing protein concentration.

Among nitrogen top dressing, there were significant differences at the 1% level in wet and dry gluten concentrations in every cultivar and breeding line. Fig. 2 shows the relationship of flour protein concentration and dry gluten concentration. Gluten concentration was a little higher in 1998 than in 1996, but almost linear and a high correlation coefficient (r = 0.97) had been recognized. Gluten concentration increased as the protein concentration increased, irrespective of cultivar, breeding line and year.

Discussion

Grain protein concentration of wheat produced in the western region of Japan tends to fall between 8 and 10% because of soil type. In order to achieve the grain protein concentration of 10–11%, which is suitable for Japanese noodles, the realistic rate of top dressing would be up to 4 gm^{-2} considering that 1 kg of nitrogen input could increase grain protein concentration by approximately 0.5%. With this amount of top dressing, the ripening stage would be delayed only by one day and such a

Table 4. Quality characteristics of each plot by top dressing at 10 days after heading

Cultivar or breeding line	Name of plot	Grain		Flour		Flour Color			Amylogram	Farinograph			Gluten		FN
		Protein	Ash	Protein	Ash	L*	a*	b*	M.V	Ab	Wk	VV	Wet	Dry	
	(gm^{-2})	(%)	(%)	(%)	(%)				(B.U.)	(%)	(B.U.)	(B.U.)	(%)	(%)	(Sec)
Chugoku 146	0	9.4	1.56	7.7	0.42	88.1	–1.80	13.9	843	57	165	27	22.7	7.5	370
	4	11.0	1.58	10.4	0.42	87.8	–1.42	13.3	860	58	145	31	30.4	10.0	351
	8	13.4	1.58	12.1	0.42	87.0	–1.40	13.1	843	60	125	36	36.7	12.0	377
ANOVA[a)]		**	*	**	ns	ns	ns	ns	ns	–	–	–	**	**	ns
Chugoku 147	0	9.1	1.60	5.6	0.46	87.7	–1.98	16.1	1,105	57	115	35	17.1	5.6	358
	4	10.8	1.63	8.6	0.45	87.4	–1.60	14.2	1,115	61	120	37	25.2	8.6	378
	8	12.4	1.63	10.6	0.45	87.5	–1.52	13.9	1,140	61	120	38	31.9	10.0	398
ANOVA		**	ns	–	–	–	–	–	–	–	–	–	–	–	ns
Shirasagikomugi	0	10.1	1.56	8.3	0.42	87.7	–1.52	13.4	950	58	155	30	23.6	7.9	373
	4	12.5	1.61	10.7	0.42	86.9	–1.26	12.9	953	61	125	37	30.8	10.0	383
	8	14.3	1.60	12.5	0.41	86.5	–1.12	12.4	933	61	120	39	38.0	13.0	388
ANOVA		**	ns	**	ns	ns	*	ns	ns	–	–	–	**	**	ns
Chikugoizumi	0	9.2	1.52	6.5	0.42	87.7	–1.78	14.3	1,130	56	120	36	18.4	6.0	351
	4	10.6	1.59	8.7	0.42	86.9	–1.56	13.8	1,075	60	125	38	25.3	8.4	309
	8	12.0	1.55	10.6	0.41	86.6	–1.41	13.1	1,060	59	120	39	31.8	11.0	290
ANOVA		**	ns	**	ns	ns	ns	*	ns	–	–	–	**	**	ns
Norin61	0	8.9	1.72	7.0	0.45	87.6	–1.54	13.8	858	58	110	37	19.4	6.4	352
	4	10.9	1.68	8.9	0.46	87.2	–1.36	13.1	885	60	110	39	26.2	8.5	356
	8	13.0	1.73	11.1	0.43	87.0	–1.17	12.6	880	60	100	41	33.6	11.0	352
ANOVA		**	ns	**	ns	ns	**	**	ns	–	–	–	**	**	ns
1CW (Reference)		14.2	1.54	13.2	0.49	87.8	–1.32	13.3	397	67	28	64	–	–	409

Milling test and FN were investigated for 1997.
Chugoku 147 and Farinograph were not analyzed by ANOVA.
*, **: Significant at the 5 and 1 % probability levels, respectively, ns: not significant.
Farinograph was investigated for 1998. FN: Falling number. a): Year was used as replications.

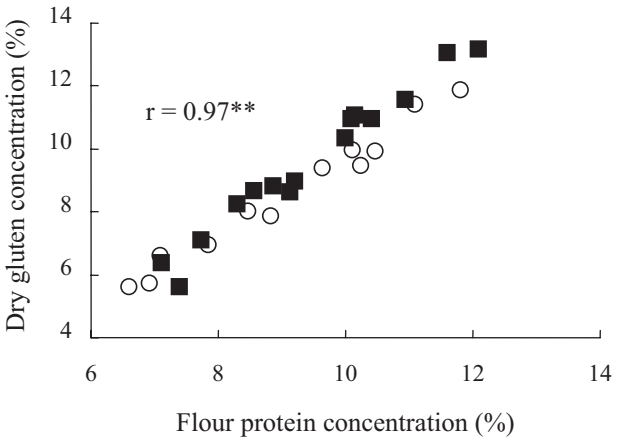

Fig. 2. The relationship between flour protein concentration and dry gluten concentration
**: Significant at the 1% probability level.
○: 1996, ■: 1998.

delay would not cause a big problem for harvesting.

The ripening stage of wheat cultivated in Kinki and Chugoku districts, a part of Western Region, is around 5 June, and since the beginning of the rainy season of the district is about 6 June in an average year, wheat can be harvested before the beginning of the rainy season. As protein concentration is an important characteristic of the wheat flour quality, harvesting date could be sacrificed for one day to obtain grains of higher protein concentration despite earlier harvesting usually being desirable.

Top dressing at inappropriate timings could cause lodging to generate immature grains. However, such an undesirable influence was not observed when nitrogen was top-dressed at 10 days after heading probably because the number of ears per plant and culm length had been determined by then. Most of the nitrogen absorbed by the wheat plants during post-anthesis phase is considered to be translocated to grains. As observed in this study, grain protein concentration increased following the

top dressing of nitrogen at 10 days after heading i.e. after anthesis. The top-dressed nitrogen, therefore, was considered to be translocated to grains.

In the field experiment where the effect of the top dressing before booting stage was examined, only a small increase of as low as 0.1–0.2% in grain protein concentration was observed with an increased nitrogen input of 1 kg[4,9]. On the other hand, Iida et al.[4] observed an increase in grain protein concentration of 0.5% with an increased input of 1 kg/10 a nitrogen at the flowering stage, which is comparable to this study where grain protein concentration increased by 0.4–0.5%. From these observations, it is considered that nitrogen top dressing from heading to three weeks after heading could increase grain protein concentration more effectively. As mentioned above, it is considered that the nitrogen top dressing after heading could increase the grain protein concentration more efficiently.

The 1,000 kernel weight and test weight increased significantly by the nitrogen top dressing. The 1,000 kernel weight and test weight were increased by the late-season top dressing[3,9], and it is thought that it can increase with late-season top dressing. Cassman et al.[2] argued that nitrogen top dressing at anthesis increases grain yield due to better filling of grains. Although, in this study, there was no significant difference in grain yield, such a possibility could not be omitted considering that both 1,000 kernel weight and test weight increased as the rate of top dressing increased.

Generally speaking, grains of higher protein concentration tend to pass a sieve and yield well. The flour yield was the highest in the 4 gm^{-2} nitrogen top dressing except for Shirasagikomugi (Chugoku 146: protein concentration 11.0%, flour yield 73.9, Chugoku 147: 10.8%, 74.5, Chikugoizumi: 10.6%, 73.6, Norin 61: 10.9%, 72.9). The flour yield was the highest at 0 gm^{-2} nitrogen top dressing after heading in Shirasagikomugi (10.0%, 74.3). When protein concentration exceeded 11%, the flour yield dropped (Chugoku 146: 72.6, Chugoku 147: 72.9, Chikugoizumi: 72.4, Norin 61: 70.8, Shirasagikomugi: 72.9, 71.7). Flour yield was the highest at protein concentration of 10 to 11%, which is suitable for Japanese noodles, and beyond the range it became low. Since the grains became hard and semolina was not fully able to be crushed using the milling conditions for soft wheat when the protein concentration increased, it was thought that the milling yield became low. The falling number became lower by nitrogen top dressing in 1997. In Chikugoizumi, the falling number decreased (304, 257, 198) as the amount of nitrogen top dressing increased in 1997, in the nitrogen top dressing block, it became less than 300. Such a phenomenon did not happen in other years, cultivars and breeding lines. Chikugoizumi sprouted due to rainfall before the ripening stage[8]. Ripening stage of Chikugoizumi was delayed for one day in the top dressing block; the delay of the ripening stage by nitrogen top dressing caused the difference in degree of pre-harvest sprouting, it is thought that the falling number dropped by the increase in the amount of top-dressed nitrogen.

This method of top dressing increased grain protein concentration and consequently gluten concentration. VV rose with increasing protein concentration on Farinograph dough properties. The protein concentration of Shirasagikomugi increased to 1CW which is similar to that of imported bread wheat (14%). However, VV (40) was far inferior to the 1CW value (64), and did not reach the suitable value for bread (60–70). A mere improvement of grain protein concentration did not make Japanese noodle cultivars suitable for bread. Nitrogen top dressing can be effective for quantitative but not for qualitative improvement of grain protein. Sato et al.[7] reported that top dressing from the snow-melting period until a week after the anthesis was effective for bread wheat cultivars. Therefore, such a way of top dressing as presented in this study is applicable not only to Japanese noodle cultivars but also to bread cultivars.

Flour color tends to deteriorate as the grain protein concentration increases. However, few data showed a significant influence of grain protein concentration on flour color. This is probably because flour color varies greatly for different years and is affected by flour yield. Negative correlations have been reported[1,3–7] between

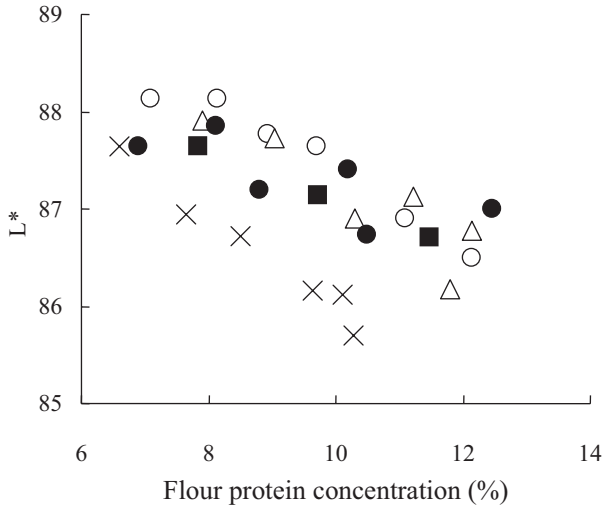

Fig. 3. The relationship between flour protein concentration and flour color (L*)

○: Chugoku 146, ■: Chugoku 147, △: Shirasagikomugi, ×: Chikugoizumi, ●: Norin 61.

grain protein concentration and flour color and therefore, color deterioration due to increased protein concentration might be unavoidable. However, Chugoku 146 is excellent in the flour color compared with the present cultivars (Fig. 3), such as Norin 61. Moreover, even when the protein concentration increased with the rates of nitrogen top dressing, within the limits of the protein concentration which is suitable for Japanese noodles, the flour color could be kept better than other cultivars and breeding lines. Thus for the cultivars of better flour color, it might be possible to minimize the degree of color deterioration by keeping the grain protein concentration within the range of 10–11% following top dressing. The mechanism of color deterioration caused by a rise in grain protein concentration is not understood and this is a future subject for investigation.

This nitrogen top dressing has been introduced into the cultivation method in Shiga Prefecture and Hiroshima Prefecture of the western region of Japan. In Hiroshima, this nitrogen top dressing for bread wheat has been put into practice, and the miller has also given a high evaluation of the result.

References

1. Barnes, P. J. (1986) The influence of wheat endosperm on flour colour grade. *J. Cereal Sci.*, **4**, 143–155.
2. Cassman, K. G. et al. (1992) Nitrogen supply effects on partitioning of dry matter and nitrogen to grain of irrigated wheat. *Crop Sci.*, **32**, 1251–1258.
3. Eguchi, H., Hirano, J. & Yoshida, H. (1996) Studies on the cultivating methods for high quality wheat in south western Japan. 2. Effect of three essential elements of fertilizer and application methods of nitrogen on quality of wheat. *Chugoku nogyoshikenjo hokoku, A*, *Sakumotsubu* (*Bull. Chugoku Natl. Agric. Exp. Stn. Crop Div.*), A**17**, 81–111 [In Japanese with English summary].
4. Iida, Y., Mitamura, T. & Ishihara, T. (1991) Influence of the soil and the cultivation conditions affect the flour color. 1 Relationship between grain protein content and flour color. *Nihon sakumotsu gakkai kiji* (*Jpn. J. Crop Sci.*), **60**(Extra issue 1), 38–39 [In Japanese].
5. Kowata, M., Watanabe, M. & Sato, A. (1996) Relationship between crude protein content and flour color of wheat cultivated in Tohoku distinct. *Tohokunoshi kenkyu shiryo* (*Misc. publ. Tohoku Natl. Agric. Exp. Stn.*) **19**, 41–45 [In Japanese].
6. Miskelly, D. M. (1984) Flour components affecting paste and noodle color. *J. Sci. Food Agric.*, **35**, 463–471.
7. Sato, A. et al. (1999) The effects of timing of nitrogen dressing on the baking quality of winter wheat. *Nihon sakumotsu gakkai kiji* (*Jpn. J. Crop Sci.*), **68**, 217–223 [In Japanese with English summary].
8. Seki, M. et al. (1999) Check of variety difference of seed dormancy in wheat. *Nihon sakumotsu gakkai kiji* (*Jpn. J. Crop Sci.*), **68**(Extra issue 2), 176–177 [In Japanese].
9. Taniguchi, Y. et al. (1999) Effect of top dressing at booting stage on crude protein content of wheat in Kyushu district. *Nihon sakumotsu gakkai kiji* (*Jpn. J. Crop Sci.*), **68**, 48–53 [In Japanese with English summary].