

Differences in Amylose Content, Amylographic Characteristics and Storage Proteins of Grains on Primary and Secondary Rachis Branches in Rice

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Abstract : The amylose content, amylographic characteristics and storage proteins of milled rice grains on the primary and secondary rachis branches were investigated. The amylose content of grains on the primary rachis branches with superior palatability was higher than that on the secondary rachis branches. Grains on the primary rachis branches showed higher maximum viscosity and breakdown values, and lower gelatinization temperature than those on the secondary rachis branches. These results suggested that the palatability of cooked rice of grains with high amylose content and amylogram values was superior to that with low amylose content and amylogram values within a cultivar under the same cultural conditions. In terms of protein fraction in the starchy endosperm, there was little difference in prolamin content and in albumin + globulin content between grains on primary and those on secondary rachis branches, whereas total protein content and glutelin content of grains on the primary rachis branches were noticeably lower than those on the secondary rachis branches for all the cultivars. Total protein, glutelin and prolamin content of a good palatability cultivar, Koshihikari were lower than those of moderate palatability cultivars Nipponbare and Reiho. These results indicate that, among storage proteins, glutelin and prolamin play important roles in rice palatability and that it is possible to use glutelin as an indicator of palatability within a single cultivar and prolamin as an indicator of palatability among cultivars.

Key words : Amylographic characteristics, Amylose, Glutelin, Palatability, Prolamin, Rachis branch, Storage proteins, Rice.

水稻における1次枝梗粒と2次枝梗粒のアミロース含有率、アミログラム特性および貯蔵タンパク質の分画の差異：松江勇次・小田原孝治・比良松道一*（福岡県農業総合試験場・九州大学農学部附属農場）

要 旨：1次枝梗粒と2次枝梗粒におけるアミロース含有率、アミログラム特性および貯蔵タンパク質の分画の差異について検討した。食味が優れている1次枝梗粒のアミロース含有率は、2次枝梗粒に比べて高かった。1次枝梗粒のアミログラム特性値は、2次枝梗粒に比べて最高粘度は高く、ブレイクダウンは大きく、糊化開始温度は低かった。したがって栽培条件が同じ場合の同一品種内ではアミロース含有率が高く、最高粘度は高く、ブレイクダウンは大きく、糊化開始温度は低い粒の方が食味は優れることが判明した。貯蔵タンパク質の分画についてみると、1次枝梗粒は2次枝梗粒に比べて、プロラミン含有率とアルブミン+グロブリンの含有率の差は明らかでなかったが、合計のタンパク質およびグルテリンの含有率は低かった。また、貯蔵タンパク質の分画の構成比率には1次枝梗粒と2次枝梗粒間で差がなかった。品種間で検討すると、良食味品種コシヒカリは日本晴、レイホウに比べて、1次枝梗粒、2次枝梗粒ともに合計のタンパク質、グルテリンおよびプロラミン含有率は低かった。これらのことから、米の食味にはグルテリンとプロラミンが重要な役割を果たしており、同一品種内ではグルテリンが、品種間ではプロラミンが食味評価の指標として使用できることが考えられた。

キーワード：アミログラム特性、アミロース、グルテリン、米、枝梗、食味、貯蔵タンパク質、プロラミン。

Rachis branches of rice, on which spikelets are differentiating, are composed of primary and secondary branches and they are the components of the panicle ear. Each spikelet is connected with a main stem through vascular bundles to transfer nutritional substances

necessary to grain development. Chaudhry and Nagato¹⁾ reported that the development of vascular bundles is not uniform but different between primary and secondary rachis branches. In addition, grains located on the primary rachis branches were superior in cooked rice palatability to those located on the secondary rachis branches and the poor palatability of the grains on the secondary rachis branches could lower the overall palatability of the whole ear as reported by Matsue et al.⁶⁾. These two facts imply that the

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accumulation of storage components in rice grains is different between grains on the primary and secondary rachis branches.

Among accumulating components in rice grains, protein and starch occupy the large quantities, and they are now widely accepted as good indicators to evaluate rice palatability. Amylose, one component of grain starch, is considered to be closely related with cooked rice stickiness and its content is used as a marker to evaluate rice palatability. On the other hand, little is known about the relationship between individual rice storage protein and palatability although whole protein content as well as amylose content in grains is the markers of rice palatability.

The present study was conducted to investigate the difference in chemical property of amylose, four kinds of storage protein and amylographic characteristics between rice grains collected from primary and from secondary rachis branches separately. The relationship between each of the chemical components and rice palatability, and possibility of improvement for rice palatability by modification of ear morphology were discussed.

Materials and Methods

Three Japanese rice cultivars with different palatability; Koshihikari (a highly palatable cultivar), Nipponbare and Reihou (a moderately palatable cultivar), were used. These cultivars were grown at the experimental field of the Buzen Branch of Fukuoka Agricultural Research Center in 1992. Seedlings of each

cultivar were transplanted in the paddy field on June 15 with a planting space of 15 cm × 30 cm (four plants per hill). The soil was clay loam (Alluvial soil). The total amount of nitrogen fertilizer application for Koshihikari, Nipponbare and Reihou was 8.5 kg, 9.5 kg and 12 kg per 10 a, respectively. The total amount of P₂O₅ and K₂O fertilizer application for Koshihikari, Nipponbare and Reihou was 6 kg, 9 kg and 12 kg per 10 a, respectively. Koshihikari plants matured in late September, Nipponbare in early October and Reihou in late October. After harvesting, the plants were solar-dried. The number of secondary rachis branches of Koshihikari, Nipponbare and Reihou were about 17, 14 and 13 per panicle, respectively. Grains were collected separately from the primary and secondary rachis branches of about 80 ears for each cultivar, and were hulled. The hulled grains more than 1.8 mm in thickness were milled at a 90% milling rate and stored at 5°C until analysis. Using hulled grains, 1000-grain weight and inspection grade were determined. Inspection grade of hulled grains was judged by the Fukuoka local food agency office under the Ministry of Agriculture, Forestry and Fisheries.

Amylose and amylography analyses were carried out from January to February, in 1993, using the methods described in the previous paper⁵⁾.

Protein fraction analysis was made with the method reported by Ogawa et al.⁸⁾ as modified by Takebe[#] in July, 1993. It was as follows:

Table 1. 1000-grain weight and inspection grade of grains on primary and secondary rachis branches.

| Cultivar | Rachis branches | 1000-grain weight (g) | Inspection grade* |
|-------------|-----------------|-----------------------|---------------------|
| Koshihikari | primary | 23.3d** | First grade-lower |
| | secondary | 21.5a | Second grade-lower |
| Nipponbare | primary | 23.4ed | First grade-middle |
| | secondary | 21.6ab | Second grade-middle |
| Reihou | primary | 23.8f | Second grade-middle |
| | secondary | 22.3c | Third grade-upper |

* Inspection grade consists of 9 grades of quality; i.e., First grade-upper, -middle, -lower, Second grade-upper, -middle, -lower and Third grade-upper, -middle, -lower.

** Means with the same letters are not significantly different by Duncan's multiple range test at 5% level.

First, 500 mg of powdered milled rice grains were homogenized in 10 ml of solution containing 50 mM KH_2PO_4 -NaOH (pH 6.8) and 0.5 M NaCl, and then centrifuged. This procedure was repeated for three times. The resulting supernatant (Fraction I) contained albumin and globulin. Next, 10 ml of 0.2 M CH_3COONa -HCl (pH 1.7) and 10 μl of 1% pepsin was added to the residue and the suspension was incubated for 60 min at 37°C. After centrifugation, the supernatant containing glutelin (Fraction II) was obtained. The

final residue (Fraction III) contained prolamins. All fractions were dissolved by Kjeldahl method, and the nitrogen content was determined by Auto Analyzer. The protein content of each fraction was calculated by multiplying nitrogen content with a protein factor 5.95.

Results

The 1000-grain weight of grains on the primary rachis branches were heavier in 1000-grain weight than those on secondary rachis branches by 1.5 g to 1.8 g, and were superior in inspection grade by 2 or 3 grades (Table 1.).

Amylose content of grains on the primary rachis branches was higher than that on the secondary rachis branches by 0.6% to 1.4% (Table 2.). Amylose content of a good palatability cultivar, Koshihikari was lower than those of moderate palatability cultivars Nipponbare and Reihou both in primary and in secondary rachis branches.

Grains on the primary rachis branches showed higher maximum viscosity and breakdown values, and lower gelatinization temperature than those on the secondary rachis branches (Table 3). Maximum viscosity and

Table 2. Amylose content of grains on primary and secondary rachis branches.

| Cultivar | Rachis branches | Amylose content (% dry weight basis) |
|-------------|-----------------|-----------------------------------------|
| Koshihikari | primary | 16.9b* \pm 0.1 |
| | secondary | 15.5a \pm 0.1 |
| Nipponbare | primary | 19.9de \pm 0.3 |
| | secondary | 19.2c \pm 0.3 |
| Reihou | primary | 20.1e \pm 0.1 |
| | secondary | 19.5cd \pm 0.1 |

* Means with the same letters are not significantly different by Duncan's multiple range test at 5% level.

Table 3. Amylographic characteristics of grains on primary and secondary rachis branches.

| Cultivar | Rachis branches | Amylographic characteristics | | |
|-------------|-----------------|------------------------------|------------------------|---------------------------------|
| | | Maximum viscosity (B.U) | Breakdown values (B.U) | Gelatinization temperature (°C) |
| Koshihikari | primary | 500 | 195 | 60.0 |
| | secondary | 470 | 132 | 69.0 |
| Nipponbare | primary | 415 | 120 | 70.5 |
| | secondary | 345 | 100 | 75.0 |
| Reihou | primary | 378 | 70 | 78.0 |
| | secondary | 318 | 68 | 79.5 |

Table 4. The distribution ratio of protein extracted from grains on primary and secondary rachis branches.

| Cultivar | Rachis branches | Distribution ratio of protein fraction (%) | | | |
|-------------|-----------------|--------------------------------------------|----------|----------|-------|
| | | Albumin + Globulin | Glutelin | Prolamin | Total |
| Koshihikari | primary | 14 | 76 | 10 | 100 |
| | secondary | 12 | 78 | 10 | 100 |
| Nipponbare | primary | 14 | 74 | 12 | 100 |
| | secondary | 13 | 76 | 11 | 100 |
| Reihou | primary | 12 | 74 | 14 | 100 |
| | secondary | 14 | 73 | 13 | 100 |

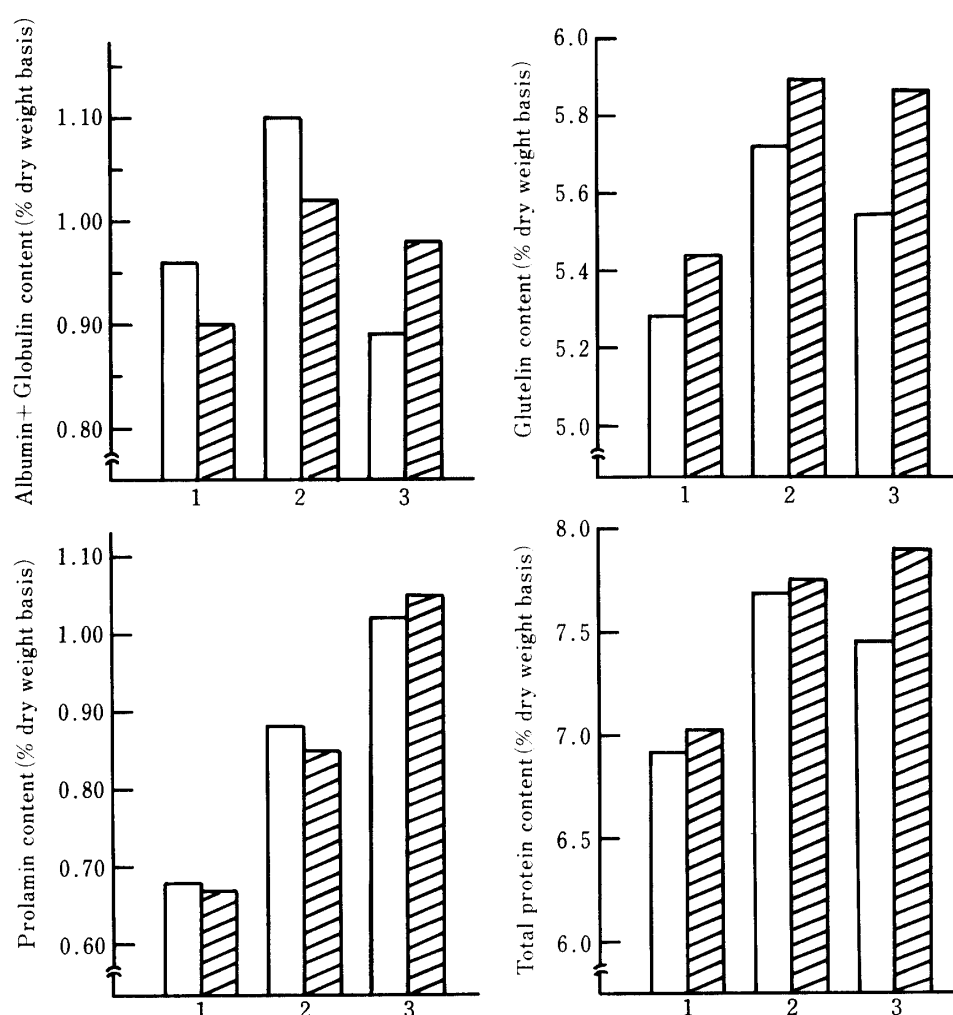


Fig. 1. Protein fractions of milled grains from primary and secondary rachis branches.
□ : Grains on primary rachis branches, ▨ : Grains on secondary rachis branches.
Cultivar names in the figure are as follows : 1, Koshihikari ; 2, Nipponbare ; 3, Reihou.

breakdown values of a good palatability cultivar, Koshihikari were higher, and gelatinization temperature was lower than those of moderate palatability cultivars Nipponbare and Reihou both in primary and in secondary rachis branches.

Among storage proteins, glutelin content was the highest, and the differences of distribution ratio of protein content between grains on primary and those on secondary rachis branches was not recognized both in primary and in secondary rachis branches for all the cultivars examined (Table 4). There was little difference in prolamin content and in albumin + globulin content between grains on the primary and those on the secondary rachis branches, whereas total protein content and glutelin content of grains on primary rachis branches was noticeably lower than those on

the secondary rachis branches for all the cultivars (Fig. 1). Total protein, glutelin and prolamin content of a good palatability cultivar, Koshihikari were lower than those of moderate palatability cultivars Nipponbare and Reihou both in primary and in secondary rachis branches. However, albumin + globulin content was not.

Discussion

The previous result of sensory test using the same materials as we used in the present study showed that grains on the primary rachis branches were superior in palatability to those on secondary rachis branches⁶⁾. In the present study, amylose content of grains on the primary rachis branches found to be higher than on the secondary rachis branches. These results suggest that amylose content shows

“positive” relationship with palatability when compared between grains on primary and those on secondary rachis branches “within” a single cultivar. This differs with the general recognition in which amylose content shows “negative” relationship with palatability when compared “among” cultivars. We do not know the reason why the result of the present study did not concordance with the general recognition, however these imply that palatability of rice cultivars should carefully be evaluated when amylose alone is used as an indicator in the evaluation of palatability.

Grains on the secondary rachis branches were inferior in amylographic characteristics to those on the primary rachis branches. Inatsu²⁾ reported that the amylogram values increased with maturity. The deterioration of amylographic characteristics for secondary rachis branches could be due to the deficiency of accumulation of starch in rice endosperm as shown by the decrease in 1000-grain weight.

In previous paper⁴⁾, maximum viscosity and breakdown values of good palatability cultivars were higher than those of ordinary cultivars. In the present study, maximum viscosity and breakdown values of grains on the primary rachis branches also showed “positive” relationship with palatability. These results suggest that maximum viscosity and breakdown values are the good indicator of palatability when compared “within” a single cultivar and “among” cultivars. Although any relationship was not found between gelatinization temperature and palatability among cultivars⁴⁾, in the present study, gelatinization temperature of grains on the primary rachis branches found to be lower than on the secondary rachis branches “within” a single cultivar. These results indicate that gelatinization temperature could be the indicator of palatability only when compared “within” a single cultivar.

Total protein and glutelin content, which was the highest among the storage proteins in primary rachis branches were lower than in secondary rachis branches. Morita⁷⁾ reported that glutelin content of a moderate palatability cultivar Nipponbare increase accordingly as total protein content increased, and Yamashita and Fujimoto⁹⁾ reported that increase in protein content by increasing nitrogen fertilization was caused by increasing glutelin con-

tent in a good palatability cultivar Sasanishiki. Prolamin content of a good palatability cultivar, Koshihikari was lower than those of moderate palatability cultivars Nipponbare and Reihou both in primary and in secondary rachis branches. Masusige et al.³⁾ reported that a ratio of prolamin content in rice starchy endosperm showed “negative” relationship with palatability among 10 cultivars. In this regard, we consider that glutelin and prolamin plays important role in rice palatability among storage proteins and that it is possible to use glutelin as an indicator for evaluation of palatability within a cultivar and prolamin as an indicator for evaluation of palatability among cultivars.

There was considerable difference between grains on primary and those on secondary rachis branches in physicochemical properties, development and appearance of the grains for all the cultivars examined. Chaudhry and Nagato¹⁾ reported that the development of vascular bundles in the primary rachis branches was better than in the secondary rachis branches. These indicate that the development of the vascular bundle in rice rachis branches greatly affect the metabolism of nutritional substances related to rice grain development and palatability.

Thus, we consider that the modification of ear morphology which decrease underdeveloped vascular bundles is a possible way to improve rice palatability.

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References

1. Chaudhry, F.M. and K. Nagato 1970. Role of vascular bundles in ripening of rice kernel in relation to the locations on panicle. *Proc. Crop Sci. Soc. Japan* 39 : 301—309.
2. Inatsu, O. 1988. Studies on improving the eating quality of Hokkaido rice. *Rep. Hokkaido Prefect. Agric. Exp. Stn. No. 66* : 1—89**.
3. Masusige, H., H. Hirai, T. Masumura and K. Tanaka 1994. Relationships between contents, distribution of protein body I, II and the cooked rice test. *Breed. Sci.* 44 (Extra issue 2) : 238*.
4. Matsue, Y., M. Yoshino and K. Harada 1989. The relation between the amylographic character-

- istics, N, Mg and K contents of milled rice and the palatability in northern Kyushu. Rep. Kyushu Br. Crop Sci. Soc. Japan 56 : 43—44*.
5. ———, K. Mizuta, K. Furuno and T. Yoshida 1991. Studies on palatability of rice grown in northern Kyushu. I. Effects of transplanting time and lodging time on palatability and physico-chemical properties of milled rice. Jpn. J. Crop Sci. 60 : 490—496***.
6. ———, K. Odahara and M. Hiramatsu 1994. Differences in protein content, amylose content and palatability in relation to locations of grains within rice panicle. Jpn. J. Crop Sci. 63 : 271—277.
7. Morita, Y. 1987. The science of rice grain. Science of cookery. 20 : 167—175****.
8. Ogawa, M., T. Kumamaru, H. Sato, N. Iwata, Z. Kasai and K. Tnaka 1987. Purification of protein body-I of rice seed and its polypeptide composition. Plant Cell Physiol. 28 : 1517—1527.
9. Yamashita, K and T. Fujimoto 1974. Studies on fertilizer and quality of rice. IV. Relationship between protein content of milled rice as affected by nitrogen fertilization and eating quality. Tohoku Agric. Exp. Stn. 48 : 91—96**.
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- * In Japanese.
** In Japanese with English summary.
*** In Japanese with English abstract.
**** Translated from Japanese by the present authors.
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