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EFFECTS OF DWARFING GENES AT THE *sd1* LOCUS ON AMYLOSE AND PROTEIN CONTENTS IN MILLED RICE

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SUMMARY

The isogenic lines for the dwarfing genes at the *sd1* locus, which originated from 'Dee-geo-woo-gen', 'Calrose76', and 'Reimei', were grown together with their common parental variety, 'Fujiminori' at the intermediate and high fertilizer levels. Another isogenic line for the dwarfing gene originating from 'Dee-geo-woo-gen' was grown together with its parental variety, Taichung 65, at a single fertilizer level. All the dwarf lines were similar to their respective parental varieties not only for amylose content but also for protein content in milled rice.

Key words: *Oryza sativa,* dwarfing gene, *sd1*, eating quality, amylose content, protein content.

The dwarfing gene originating from the Taiwanese variety 'Dee-geo-woo-gen' viz. *sd1*, and allele(s) at the *sd1* locus contributed to the development of short- culm and lodging-resistant varieties not only in Southeast Asia, but also in USA and Japan (Futsuhara, 1968; Mackill and Rutger, 1979; Kikuchi et al., 1985; Murai and Yamamoto, 2001).

For Japanese rice varieties, there is a tendency that the lower the amylose content in milled rice, the better the taste of cooked rice. For example, an excellent-taste variety 'Koshihikari' produces grains of low amylose content (Kurasawa, 1972; Inatsu, 1988). Low eating quality of most conventional Chinese *japonica* varieties is principally due to their high amylose content (Cui et al., 1999 a,b; Ise et al., 2000). In addition, high protein content in milled rice is apt to lead to low eating quality (Yamashita and Fujimoto, 1974; Inatsu, 1988; Cui et al., 1999 a,b; Ise et al., 2000).

In this study, it was investigated whether dwarfing genes at the *sd1* locus affected the amylose and protein contents or not by using isogenic lines and their parental varieties, 'Fujiminori' and Taichung 65.

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MATERIALS AND METHODS

Designation of dwarfing genes

'Calrose 76' is a short-culm mutant from 'Calrose', induced by gamma-ray irradiation in California (Mackill and Rutger, 1979). Tanisaka et al. (1994) tentatively designated the dwarfing gene of 'Calrose 76' as *sd1-c*. Further, Murai and Yamamoto (2001) used *sd1-d* and *sd1-r* as the tentative names of the dwarfing genes from 'Dee-geo-woo-gen' and 'Reimei', respectively, since *sd1-r* is an allele at the *sd1* locus. The effects of the genes on culm elongation can be expressed as follows: $sd1\text{-}d \leqq sd1\text{-}c < sd1\text{-}r < Sd1$ (short < long) (Murai and Yamamoto, 2001).

Isogenic lines of 'Fujiminori'

'Fujiminori', denoted as "F", is a tall variety which was widely grown in northern Japan during the 1960s. Two isogenic lines of the recurrent parent F were used (Murai and Yamamoto, 2001). The isogenic line for *sd1-c,* developed after eight backcrosses, was denoted as "D^c". The isogenic line for *sd1-d* ("D^d") was developed after 11 backcrosses. 'Reimei' ("R") is a gamma-ray induced mutant from F (Futsuhara, 1968). D^d, D^c, R and F were grown in Aomori Agricultural Experiment Station. Two paddy fields were fertilized at 10 and 16 g/m^2, respectively, for each of N, P_2O_5 and K_2O as total (basal + top dressings), viz. intermediate and high fertilizer levels. Thirty-nine-day-old seedlings of the four lines were transplanted to the two paddy fields with three or four seedling per hill at a spacing of 30.3×13.6 cm, on May 22, 1998. The randomized block design with two replicates was applied for the four lines in each of the two paddy fields. Amylose content of milled rice in each plot was measured by means of AutoAnalyzer® II (Bran+Luebbe K.K., Japan), the determination of which was based on coloration of amylose-iodine complex (Inatsu, 1988). Protein content in milled rice of each plot was measured on the basis of near infrared spectroscopy by means of InfraAlyzer® 450 (Bran+Luebbe K.K., Japan).

Isogenic line of 'Taichung 65'

Another isogenic line for *sd1-d,* which was developed after ten backcrosses with a *japonica* variety, Taichung 65 (Murai et al., 1995), was used. This line was denoted by "T^d". Taichung 65 ("T65") and T^d were grown at a fertilizer level of 8 g/m^2 for each of N, P_2O_5 and K_2O as total (basal + top dressings), with three replicates. Twenty-six-day-old seedlings were transplanted to a paddy field at the Faculty of Agriculture, Kochi University with two seedlings per hill at a spacing of 30.0×15.0 cm, on June 1, 2000. To measure amylose content in milled rice in this experiment, the method of Juliano (1971) was used. This was based on coloration of amylose-iodine complex. Protein content in milled rice for each plot was measured on the basis of near infrared spectroscopy by means of Near-infrared reflectance® Model 5000 (FOSS NIR Systems, Inc., USA).

RESULTS AND DISCUSSION

Table 1 shows the results of the analysis of variance for data on D^d, D^c, R and F at two fertilizer levels. Regarding amylose content, the effect of fertilizer level was significant at the 5% level: each of the lines showed small or no decrease from the intermediate to high fertilizer levels (Table 2). These decreases were smaller than the LSD at the 5 % level (Table 2), indicating nonsignificant fertilizer response of each line. At each fertilizer level, the amylose contents of the four lines were similar to one another (Table 2), which was supported by nonsignificant effect of lines for this trait (Table 1). Regarding protein content, the effect of fertilizer level was significant (Table 1). Indeed, each line showed significant increase from the intermediate to high fertilizer levels (Table 2). At the high fertilizer level, all the four lines were similar to one another. At the intermediate fertilizer level, only D^d was significantly higher than F, whereas the other three lines including F were similar to one another.

Table 1. Analysis of variance for amylose and protein contents of D^d, D^c, R and F ('Fujiminori') at the two fertilizer levels. Numerals in the table exhibit F-values for effects of fertilizer levels, lines, and the interaction between them in each trait.

	Source of Variation		
Trait	Fertilizer levels	Lines	Interaction (F.×L.)
Amylose (%)	8.00*	< 1	< 1
Protein (%)	65.05**	5.15*	1.10

Degrees of freedom for fertilizer levels, lines, the interaction, blocks and error are 1, 3, 3, 1 and 6, respectively. Sum of squares for the interaction between lines and blocks in the intermediate fertilizer level, and that in the high fertilizer level were pooled. It was used as the error sum of squares in the present study (according to the report of Okuno et al. (1971)).
*,**Significant at 5 and 1% levels, respectively.

Table 2. Amylose and protein contents of D^d, D^c, R and F ('Fujiminori') at the two fertilizer levels.

Trait	Fertilizer level	D^d	D^c	R	F	$LSD_{(5\%)}$ [1]
Amylose (%)	Intermediate	20.3	20.6	20.6	20.4	0.5
	High	20.3	20.2	20.2	20.0	
Protein (%)	Intermediate	7.3**	6.7	6.7	6.5	0.5
	High	7.8	7.5	7.8	7.5	

[1] Calculated from error mean square of analysis of variance (Table 1)
**Difference from F is significant at the 1% level.

The amylose content of T^d (14.6%) was similar to that of T65 (14.9%). The protein content of T^d (6.6%) was similar to that of T65 (6.9%). Analysis of variance for the two traits indicated no significant differences between the two lines.

Results of the present study indicate that *sd1-d, sd1-c* and *sd1-r* have no pleiotropic effects on amylose content. Regarding protein content, all the dwarf lines were similar to their respective parental varieties, with the exception of D^d at the intermediate fertilizer level. This indicates none or small pleiotropic effects of the three dwarfing genes on this trait. Hence, it is suggested that *sd1-d, sd1-c* and *sd1-r* exert no or small effects on eating quality. Factors other than amylose and protein contents, *e. g.*, tenderness and scent affect eating quality. To reach a final conclusion, sensory test for cooked rice may be necessary.

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