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Evidence for maternal effect in the inheritance of grain protein in crosses between cultivated and wild tetraploid wheats

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Summary. Reciprocal crosses were made between cultivated wheat (*Triticum turgidum* var. 'durum') and a high-protein line of wild tetraploid wheat (*T. turgidum* var. 'dicoccoides'). F_1 grains (on maternal spikes) were very similar to the selfed grains on the maternal parent in protein percentage, weight and protein content. These traits were also analyzed in F_3 grains developed on F_2 spikes of segregating populations derived from reciprocal crosses between the same cultivated parent and another high-protein line of var. 'dicoccoides'. No significant differences in the mean values of these traits were found between the reciprocal crosses, indicating no cytoplasmic effect. It has been concluded that these grain characteristics are largely determined by the maternal plant.

Key words: Wheat – *Triticum* – Grain protein – Grain weight – Maternal effect – Cytoplasmic effect

Introduction

The inheritance of grain protein percentage in wheat has been a subject of intensive studies in recent years (Diehl et al. 1978; Cowley and Wells 1980; Kraljević-Balalić et al. 1982; Löffler and Busch 1982). The accumulating data indicate that in common wheat grain protein percentage is highly heritable and is determined, in most cases, by several genes (Johnson et al. 1968). However, differences in grain protein percentage between certain varieties of common wheat have been found to be determined by only one or two genes (Haunold et al. 1962; Cowley and Wells 1980). The genes for low grain protein percentage show a weak dominance over those for high protein (Chapman and McNeal 1970; Johnson et al. 1973; Diehl et al. 1978; Halloran 1981) though in some cases evidence has been obtained for the opposite (Hsu and Sosulski 1969; Cowley and Wells 1980).

The above mentioned studies were carried out in cultivated varieties, which exhibit a relatively narrow range of grain protein percentage. These studies also lack data on protein determination in F_1 grains.

Recently Avivi (1978, 1979) found that several collections of wild tetraploid wheat *Triticum turgidum* var. 'dicoccoides' contained an exceptionally high percentage of grain protein (24–29%) – twice as much as that of cultivated wheats. Little is known about the inheritance of this trait. Avivi et al. (1983) and Levy and Feldman (1983) obtained evidence indicating that several different genes not found in cultivated wheat are involved in the determination of this trait. The genes for low grain protein are partially dominant over those for high grain protein.

The aim of this study was to evaluate the maternal contribution to grain protein. The analysis was carried out in hybrids and segregating populations of reciprocal crosses between cultivated and high-protein wild tetraploid wheats.

Materials and methods

Plants of cv. 'Inbar' of 'durum' wheat (*Triticum turgidum* var. 'durum') containing about 14% protein in their grains and those of a high-protein line of wild wheat, *T. turgidum* var. 'dicoccoides' (TTD15) containing about 29% grain protein were grown individually in five liter pots filled with a mixture of 50% volcanic gravel, 33% peat and 17% vermiculite (v/v). The peat was enriched with basic fertilizer containing 10% pure nitrogen. This supplied about 150 mg pure nitrogen per pot. In addition, 11 g of a slow-release fertilizer added to the mixture were alloted to each pot. This was calculated to release about 1,500 mg of pure nitrogen per pot within a period of three to four months.

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Selfing and reciprocal crosses between 'Inbar' and TTD15 were carried out on the same spike as following: the emasculated two basal florets in the central part of the spike were either selfed or crossed by the method described by Millet and Pinthus (1980). Comparison of crossed and selfed grains was made on the same spike. Because of expected spike fragility in TTD15, each spikelet was tagged at the time of pollination to ensure identification of selfed and crossed grains.

The nature of the products was verified by comparing the electrophoretic pattern of the total endosperm proteins in SDS polyacrylamide gel electrophoresis as described by Galili and Feldman (1983). No contamination with undesirable pollen was detected in the derived grains.

The grains of each half of the spike (either selfed or crossed) were pooled, weighed and their protein percentage was determined by the micro-Kjeldahl method.

F_2 populations of the reciprocal crosses between another high-protein line of var. 'dicoccoides' (line TTD28) and 'Inbar', as well as both parents, were grown in a sandy soil and fertilized with 12 g of pure nitrogen per m^2. Plants were spaced 20 cm apart within a row and 60 cm between rows. Protein percentage was determined collectively on a sample of 20–30 grains from each F_2 plant by infrared reflectance analysis using a Neotec GQA grain quality analyzer.

Results

F_1 and selfed populations

Although the values of protein percentage were relatively high in this experiment, grains developed on TTD15 spikes had considerably and significantly higher protein percentage than those developed on 'Inbar' spikes (Table 1). However, in both lines, the percentages of protein in the grains derived from either selfing or crossing the florets on the same spike were very similar. Comparison of crossed and selfed grains on the same spike eliminated the 'between spikes' component of the environmental variance. Eliminating this factor, which was found to be the major environmental variance component (Table 1), resulted in a more precise estimation of the genetic influence. Nevertheless a slightly but not significantly higher protein percentage was found in grains derived from TTD15 as male parent.

Grain weight followed a pattern similar to that of protein percentage. The two maternal parents differed in this trait, TTD15 having considerable and significant lower values (Table 1). This difference was maintained in the crossed and selfed grains regardless of the pollen source. Nevertheless, crossed florets yielded grains somewhat, but nonsignificantly, heavier than selfed florets on the same spike (Table 1).

Grains developed on spikes of var. 'dicoccoides' contained significantly lower amount of protein than those which developed on the spikes of 'Inbar' regardless of the pollen source (Table 1). This is mainly due to the larger grain weight of 'Inbar' while the higher protein percentage of TTD15 was insufficient to compensate for its smaller grains. Crossing the florets rather than selfing them, resulted in a nonsignificant increase of about 6% in grain protein content. In each of the four combinations, protein content of the grains was strongly correlated with their weight. Correlation coefficients ranged from 0.81 to 0.97 for the environmental effects and were always significant.

F_3 grains of F_2 segregating populations

The two F_2 populations from the reciprocal crosses between 'Inbar' and TTD28 did not differ significantly in mean protein percentage, weight and protein content of their grains (Table 2). Compared with the parental

Table 2. Means ± SE of protein percentage, weight (mg) and protein content (mg) of grains of 'durum' wheat (cv. 'Inbar'), var. 'dicoccoides' (line TTD28) and of F_3 grains, developed on F_2 plants from the two reciprocal crosses. (N = number of plants in each group)

	Parents		F_2 populations	
	'Inbar' ($N=18$)	TTD 28 ($N=7$)	'Inbar' × TTD 28 ($N=190$)	TTD 28 × 'Inbar' ($N=220$)
Protein percentage	13.4±0.3	27.5±0.8	18.8±0.2	19.2±0.2
Grain wt	63.0±4.5	23.1±1.1	49.2±0.8	50.1±0.8
Protein content	8.5±0.6	6.3±0.4	9.1±0.2	9.5±0.2

Table 1. Protein percentage, weight (mg), and protein content (mg) of grains developed on one side of the spike of var. 'dicoccoides' (line TTD15) of 'durum' wheat (cv. 'Inbar'), derived from either selfing the florets or crossing them with the other parent

	Maternal parent TTD 15			Maternal parent 'Inbar'			Environmental variance	
	Pollinator TTD 15	Pollinator 'Inbar'	SE*	Pollinator TTD 15	Pollinator 'Inbar'	SE*	Between spikes	Within spike
Protein percentage	29.5	29.0	0.3	19.8	19.4	0.3	15.65	0.56
Grain wt	15.6	16.8	1.5	48.5	47.1	1.3	128.58	11.74
Protein content	4.5	4.7	0.3	9.6	9.0	0.3	5.74	0.59

* SE of the difference between pollinators

protein percentage, the mean values of the F_2 populations were slightly lower than the mid-parents' values, indicating that the genes for low protein of the cultivated parent show a weak dominance over the high-protein genes of the wild parent. On the other hand, the mean values of weight and protein content of the grains of F_2 populations were higher than the mid-parental values, indicating dominance of genes for large grain weight.

The analysis of the F_3 grains data yielded the following highly significant phenotypic correlation coefficients: 0.845 between grain weight and protein content, 0.164 between protein percentage and protein content and −0.366 between protein percentage and grain weight.

Discussion

Protein percentage, weight and protein content of F_1 grains from reciprocal crosses between 'durum' wheat and high-protein var. 'dicoccoides' were determined by the maternal parent. Only small and nonsignificant differences were detected for these grain characteristics between crossed and selfed grains, while large differences were found between grains of reciprocal crosses. Only a slight and nonsignificant paternal effect on grain protein percentage was noticed, while F_1 grains derived from reciprocal crosses had a slight and nonsignificant advantage over selfed grains of the same maternal parent in weight and protein content.

The overwhelming influence of the maternal parent on both protein percentage and grain weight can be ascribed either to the endosperm genetic constitution, to cytoplasmic inheritance or to maternal effect. The first of these possibilities assumes that grain protein level and grain size are determined, at large, by the duplicated genetic contribution of the maternal parent to the triploid endosperm. Although this assumption seems implausible, it should be further examined by analyzing these characteristics in F_2 grains. In fact, Millet and Pinthus (1980) examined grain weight in F_2 populations (developed on F_1 spikes) and rejected the possibility that grain weight was influenced by the endospermic genetic constitution.

The comparison between the two F_2 populations of reciprocal crosses between 'Inbar' and high-protein var. 'dicoccoides', revealed no cytoplasmic effect on grain characteristics. Similar results have been obtained for grain protein percentage by McNeal et al. (1968) in crosses between common wheat cultivars differing in their protein percentage. However, different results were obtained by Worstell (1982) who reported that grain protein percentage of F_2 populations in crosses between the synthetic amphiploid *T. turgidum* var. 'dicoccoides' – *Ae. squarrosa* and common wheat (cv. 'Newton') were dependent on the direction of the cross.

The most plausible explanation for the reported results is that the analyzed grain characteristics are largely determined by the maternal tissues, as has been previously suggested for grain weight (Millet and Pinthus 1980). This assumption fully accounts for the observed differences in the F_1 grain characteristics in the reciprocal crosses and the lack of differences between the selfed and crossed grains on the same maternal plant. Support for this assumption comes from physiological studies carried out in cultured spikes in which it was unequivocally demonstrated that grain protein percentage is controlled by the source level rather than by the sink level (Donovan and Lee 1978; Donovan 1979).

Different maternal tissues and mechanisms may play a role in determining grain protein percentage and grain weight. Elucidating the nature of the maternal effect requires the manipulation of plant tissues and environmental conditions. Millet and Pinthus (1984) found that when the source is not a limiting factor in grain filling, floral organs that have a morphogenetic effect on grain size determine grain weight. On the other hand, the effect of the maternal plant on grain protein percentage is presumably exerted through a different mechanism. Preliminary studies (Millet, unpublished) indicate that grain protein percentage was almost unaffected by changing the relative size of the sink but responded to soil nitrogen level. These results and those of Donovan and Lee (1978) show that the source rather than the sink determines the percentage of grain protein. It is tempting to speculate therefore, that grain protein percentage is affected by the ratio of amino acids to sugars in the green tissues or by their relative translocation to the developing grains, while grain protein content is largely determined by the storage capacity of assimilates in the grain. This storage capacity (grain size) is maternally affected.

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