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Polymorphism of the glutenins and gliadins in emmer wheat from Spain

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SUMMARY – Emmer wheat (*Triticum turgidum* ssp. *dicoccum* Schrank) is hulled wheat that survives in marginal areas in the Mediterranean region and that could be used as source of genes for wheat improvement. The storage protein composition of ninety-eight accessions of emmer wheat from Spain have been analysed by SDS-PAGE (HMW- and LMW-Gs) and A-PAGE (gliadins). For the HMW-Gs, four allelic variants were detected for the *Glu-A1* locus; one of them has not previously been described. For the *Glu-B1* locus, three of nine alleles detected have not been found to date. A high degree of variation has been found for the *Glu-3* (LMW-Gs) and *Gli-1* loci (γ- and ω-gliadins).

Key words: Diversity, electrophoresis, germplasm, hulled wheat.

RESUME – *"Polymorphisme des gluténines et des gliadines chez l'amidonnier en Espagne". L'amidonnier (*Triticum turgidum *ssp.* dicoccum *Schrank) est un blé couvert qui survit dans les aires marginales de la région méditerranéenne et qui peut être utilisé comme source de gènes pour l'amélioration du blé. La composition protéique de stockage de quatre-vingt-dix-huit accessions d'amidonnier provenant d'Espagne a été analysée par SDS-PAGE (HMW- et LMW-Gs) et A-PAGE (gliadines). Pour le HMW-Gs, quatre variants alléliques ont été detectés, pour le locus* Glu-A1, *l'un d'eux n'a pas été précédemment décrit. Pour le locus* Glu-B1, *trois des neuf allèles détectés n'ont pas été trouvés jusqu'à présent. Un haut degré de variation a été trouvé pour les loci* Glu-3 *(LMW-Gs) et* Gli-1 *(γ- et ω-gliadines).*

Mots-clés : *Diversité, électrophorèse, germoplasme, blé couvert.*

Introduction

First studies of the molecular aspects of quality in the world collections of wheat revealed low variability for some protein components with sharp influence on bread making performance (Shewry *et al.,* 1989). For this reason, the search for species that could be useful in contributing genes for quality improvement has great importance in most wheat breeding programs (Jauhar, 1993). One of these species is emmer wheat, *Triticum turgidum* ssp *dicoccum* Schrank.

Emmer wheat is primitive, hulled wheat, allopolypoid species with the genome formula **AABB,** which was widely cultivated in the past under the name of "farrum". At present, this specie survives as a crop in marginal areas in the Mediterranean region where it is used for feed livestock (D'Antuono, 1989). Nevertheless, the interest toward hulled wheats has been increasing again over the last few years. This increased interest is due to the low-input techniques used for their management (D'Antuono, 1989), the increasing demand for unconventional foods, and the therapeutic properties attributed to their derivatives (Auricchio *et al*., 1982). On the other hand, these species have shown potential as a source of useful genes for breeding naked wheats (Sharma *et al*., 1981; Srivatava and Damania, 1989).

The aim of this study was to evaluate the polymorphism of seed storage protein present in emmer wheat accessions from Spain.

Materials and methods

Seed samples for 98 accessions of emmer wheat used in this study were obtained from Centro de Recursos Fitogenéticos INIA (Alcalá de Henares, Spain) and National Small Grain Collections

(Arberdeen, USA). In order to compare, emmer wheat accessions contained standard allelic variants for *Glu-A1* and *Glu-B1* loci described by Vallega and Waines (1987) were used.

Seeds crushed into a fine powder were used to extract the endosperm storage proteins. Gliadins were extracted with a 1.5 M dimethylformamide aqueous solution and fractionated by acid-polyacrylamide gel electrophoresis (A-PAGE) at 8% (C: 2.67%), according to Khan *et al.* (1985).

Glutenins were extracted according to Singh *et al.* (1991) and fractionated in vertical SDS-PAGE slabs at a polyacrylamide concentrations of 8 and 10% (w/v, C: 1.28%) with and without 4 M urea according to Lafiandra *et al.* (1993). Electrophoresis was performed at a constant current of 30 mA/gel at 18ºC. Gels were stained overnight with 12% (w/v) trichloroacetic acid solution containing 5% (v/v) ethanol and 0.05% (w/v) Coomassie Brilliant Blue R-250. Destaining was carried out with tap water.

Results and discussion

Up to thirteen allelic variants (four alleles at *Glu-A1* locus and nine at *Glu-B1* locus) were found between the evaluated lines. Comparing their mobilities to those previously found in emmer wheat we identified new allelic variants, one at the *Glu-A1* locus and three at the *Glu-B1* locus. These new alleles were designated with progressive Roman numerals according to the nomenclature of Vallega and Waines (1987), who identified alleles *Glu-A1-I* to *IV*, and *Glu-B1-I* to *VI*. Therefore, the new alleles found in the present work were named *Glu-A1-V* and *Glu-B1-VII*, *VIII* and *IX* (Table 1).

Table 1. Allele frequencies at *Glu-1* loci in 98 Spanish accessions of emmer wheat

Locus	Allele	HMW glutenin subunits	Accessions	
			N_	%
Glu-A1	a	1	87	88.78
	c	Null	7	7.14
	j	III (one)	3	3.06
	Not allocated	V (one)	1	1.02
Glu-B1	b	7+8	72	73.48
	d	6+8	5	5.10
	n	II (two)	9	9.18
	p	IV (two)	1	1.02
	q	V (one)	1	1.02
	r	VI (one)	1	1.02
	Not allocated	VII (one)	7	7.14
	Not allocated	VIII (two)	1	1.02
	Not allocated	IX (two)	1	1.02

Novel subunit V has a very fast migration, being ever much faster that subunit 2* (*Glu-A1b*) of the bread wheat.

For the *Glu-B1* locus, new subunit VII presents only one major subunit. In 8% gels was found lightly faster moving than subunit VI, being too faster in urea gels. Likewise, subunit VIII showed only one band with the same mobility as subunit VI. However, when it was analysed in urea gels, it was observed that while subunit VI presented a single band, two components with very different migration rate were detected for the new allele VIII. In 8% gels without urea, the *x* subunit of *Glu-B1* IX has the same mobility as *Glu-Bx*-7, whereas the γ subunit migrate at the same rate as *Glu-B1* VI subunit.

Although many of the studied accessions presented similar patterns for HMW-Gs, they were rich in variability for LMW-Gs and gliadins. In fact when the LMW-Gs were analysed, eleven alleles for *Glu-A3* locus and twelve for *Glu-B3* locus were identified. Twenty-one different patterns were found considering the variation at both loci. For the gliadins, thirty-six patterns were detected, taking into account the whole variation for *Gli-1* and *Gli-2* loci (Fig. 1).

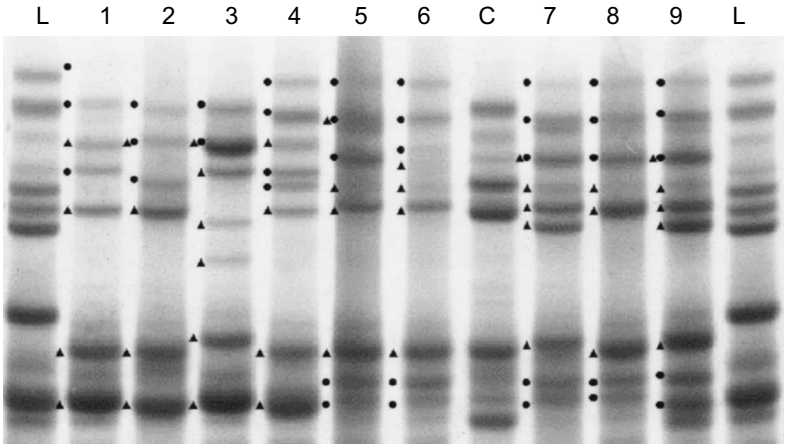

Fig. 1. A-PAGE of gliadins from emmer wheats and standard durum wheats (L = Langdon, C = Claro de Badajoz). Gliadins encoded at the *Gli-A1* locus (●) and at the *Gli-B1* locus (▲) are shown.

In conclusion, because of the high variation detected for the endosperm storage proteins of emmer wheat, we think that this specie could be used as source of genes for quality improvement in durum wheat.

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