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Isozyme variation in some populations of wild diploid wheats in Iran

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Abstract

Isozyme electrophoresis data of seed extracts from 11 populations of diploid wheat species (*Triticum boeoticum* Bioss. and *Triticum urartu* Thumanian ex Gandilyan), distributed mainly in the western and west-northern Iran, were investigated. The five enzyme systems used were peroxidase, polyphenol oxidase, superoxide dismutase, malate dehydrogenase and catalase. The first three were found to be useful as molecular marker for characterization of diploid wheat populations. A total of 13 bands from three enzyme systems were recorded. The value of a 'Jaccard's' similarity coefficient ranges from 0.333 to 1.000. Data analysis was done using clustering method UPGMA. On the basis of Jaccard's coefficient, the obtained dendrogram supports previous relationship between *T. boeoticum* and *T. urartu* as separate species as well as reflecting their distinct gene pools and substantiating their specific recognition despite the overall morphological similarity.

Keywords: Diploid wheat; Isozyme variation; Genetic relationships; *Triticum*

1. Introduction

The genus *Triticum* L. is composed of diploid, tetraploid and hexaploid species including both domesticated and wild species. Diploid wheats are genetically the most basic wheat types with a diploid chromosome number ($2n = 14$) (Loje et al., 2003). Diploid *Triticum* species are composed of three species: the cultivated *Triticum monococcum* L., its immediate wild relative *Triticum boeoticum* Boiss. and the wild *Triticum urartu* Thumanian ex Gandilyan which are the A genome donors of polyploid wheats (Gill and Kimber, 1974; Chapman et al., 1976; Konarev, 1983; Morris and Sears, 1967). Johnson (1975) suggested that, according to the protein electrophoretic analysis, *T. urartu* could be the B genome donor of polyploid wheats. The classification of these species has been carried out mainly on the basis of morphological characters.

Domesticated diploid wheat *T. monococcum* var. *monococcum* is still cultivated in mountain areas south-eastern of Europe and Turkey (Harlan, 1981). *T. boeoticum* is wildly distributed throughout the eastern Mediterranean countries like Armenia, Azerbaijan, Bulgaria, Iran, Iraq, Lebanon, Syria and Turkey (Johnson, 1975). *T. urartu* is restricted

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mainly to the Fertile Crescent and distributed in Armenia, Azerbaijan, Iran, Iraq, Lebanon, Syria and Turkey (Johnson, 1975). The populations of diploid wheats are distributed mainly in the western and west-northern areas of Iran.

The characterization of plant germplasm has traditionally been carried out through the study of morphological traits. However, currently the use of molecular techniques has become widespread in the study of biological characters (Crawford, 1990; Weising et al., 1995). Isozyme electrophoresis, being one of molecular techniques, has proven to be highly useful as biochemical markers for solving various problems of plant taxonomy in order to distinguish or to confirm species (alpha systematics). The characterization also allows us to measure divergence among populations at any level from within and among species of related genera (beta systematic or phylogenetics) (Vanijajiva et al., 2003). Knowing about the nature of genetic variability in diploid wheats is important because of its potential use in wheat breeding (Gale and Miller, 1987; Appels and Lagudah, 1990; Szabo and Hammer, 1996).

The aims of the study were to: (1) describe electrophoretic isozyme phenotypes and their variation patterns among the populations of wild *Triticum* species (*T. boeoticum* and *T. urartu*), (2) evaluate possibilities to use allozymes as molecular characters to discriminate between the *Triticum* species and populations and (3) estimate phylogenetic affinities and relative extent of allozyme divergence between the *Triticum* species by cladistic and phenetic analyses of allozyme diversity. The present work is the first report on isozyme variation in Iranian diploid wheats.

2. Material and methods

2.1. Plant materials

Seeds of some populations of *T. boeoticum* and *T. urartu* were collected from different regions of western and west-northern Iran during 1996–2000 by A. Salimi. Botanical identification was determined in accordance with Dorofeev et al. (1979) and Gandilyan (1980) monographs. All of the specimens were deposited in the Central Herbarium of Tarbiat Moallem University in Tehran. Taxonomic and locality data of the taxa used for the experiment are listed in Table 1. The distribution map of the collected plants is presented in Fig. 1.

In this study, seeds were germinated and grown in field in the University of Tehran (November 2004 to May 2005) and then seeds produced by these plants were used for electrophoretic analysis. Every time a single seed from each population was used.

2.2. Isozyme analysis

For protein extraction, single dry seeds (endosperm and embryo) were powdered. The flour of seeds were homogenized in the extraction buffer (Tris–Gly 0.001 M, pH 7.2 containing glycerol 0.1 v/v) 1:6 (w/v) and centrifuged (13,000 rpm at 4 °C) for 60 min. The supernatant was collected and stored at −20 °C until was used for analysis. Because of its high resolution (Soltis and Soltis, 1990), native polyacrylamide gel electrophoresis (PAGE) was performed according to a modified method of Davis (1964) using vertical slab gels (1 mm thick) and was set up forming a discontinuous system of two layers: (i) resolving gel: 8 cm layer of 15% polyacrylamide and (ii) stacking gel: 3 cm layer

Table 1
Taxonomic and locality data of diploid wheat populations used in the study

Species	Population symbols	Locations
Triticum boeoticum	B1	Ivan–Islamabad
	B2	Oshnavie–Piranshahr
	B3	Oshnavie–Piranshahr
	B4	Oshnavie–Piranshahr
	B5	Taleghan valley
	B6	Bane–Marivan
	B7	Khoramabad
	B8	Khoramabad
Triticum urartu	U1	Sardasht
	U2	Kermanshah
	U3	Sanandaj