## Poster Abstract - B.27

## CLONING AND CHARACTERIZATION OF MADS BOX GENES IN WHEAT

## A.R. PAOLACCI\*, S. VAROTTO\*\*, O.A. TANZARELLA\*, E. PORCEDDU\*, M. CIAFFI\*

\*) Dipartimento di Agrobiologia e Agrochimica, University of Tuscia, Via S. Camillo de Lellis, 01100 Viterbo, Italy – ciaffi@unitus.it \*\*) Dip. di Agronomia Ambient. e Prod. Veget., Univ. Padova, Via Romea 16, 35020 Legnaro (PD)

## MADS-box genes, flower development, phylogenesis, Triticum

The genes of the MADS-box family encode transcription factors which play key roles in the control of development and of signal transduction in eukaryotes. The MADS family has been partitioned into two main lineages, type I and type II, both are represented in plants, animals and fungi. Type II genes have been subjected to extensive researches, whereas much less is known on type I genes in plants. Type II MADS proteins of plants, or MIKC-type, are involved in flower and ovule morphogenesis, vegetative growth, embryogenesis and fruit formation. Through phylogenetic analysis based on sequence comparison, they have been classified into 13 subfamilies. The best studied MIKC transcription factors are those controlling the determination of floral organ identity. Analysis of homeotic floral mutants in Arabidopsis and Antirrhinum resulted in the formulation of the ABC genetic model, which explains how the combined functions of three classes of genes (A, B and C) determine the identity of the organs (sepals, petals, stamens and carpels) of the four concentric floral whorls. The ABC model was then extended to include two more functions, yielding an "ABCDE model". Understanding of the molecular control of flower development in grasses and other monocots lags behind that in Arabidopsis and other eudicots. In spite of the substantial differences of the typical flower structure in monocots and dicots, recent studies, namely in rice and maize, have shown a significant conservation of floral homeotic gene functions between monocots and dicots. On the basis of these observations, it has been suggested that the ABC model can essentially be extended to monocots. In wheat little is known on the molecular basis of flower development, in fact only five MADS-box genes of the AP1, AP3, PI, AGL6 and AG groups had been isolated before the present research.

In the present research 20 cDNA sequences (WM) of wheat MADS-box genes have been cloned following two different approaches. Eight sequences were identified and cloned using a method of differential analysis, whereas twelve more sequences were identified in the EST database and cloned through RT-PCR of mRNA from fully emerged wheat spikes. Full-length cDNA sequences were obtained by RACE extension of some of the cloned fragments and their expression patterns were studied by RT-PCR in different plant and floral tissues. A phylogeny reconstruction of 78 MADS box genes was carried out in order to determine the evolutionary relationships between the isolated wheat cDNA sequences and other known plant MADS box genes. The phylogenetic tree showed that 14 of the wheat clones could be assigned to 8 distinct subclasses of the MADS gene family, whereas three (WM16, WM17 and WM18) did not cluster with any of the known subclasses, a condition resembling that of other monocot MADSbox genes described as solitary sequences or orphan genes. The phylogenetic tree indicated that we were able to clone wheat orthologs representative of most, if not all, dicot MIKC-type genes controlling the floral homeotic functions A, B, C, D and E. The expression patterns of the isolated sequences were compared with those of functionally characterized MADS-box genes from Arabidopsis and monocot species. Sequence similarity and comparable expression patterns were the parameters used for a preliminary prediction of the potential functions of the genes corresponding to the isolated wheat MADS-

box sequences. It is now in progress the expression analysis of the MADS clones in wheat floral organs by *in situ* hybridization.