

UNRAVELLING *KNOTS* IN PEACH: MAPPING AND FUNCTION OF *KNOPE1*, A CLASS I *KNOTTED-LIKE* GENE ENCODING A HOMEODOMAIN CONTAINING TRANSCRIPTION FACTOR

G. TESTONE*, E. CONDELLO*, C. NICOLODI*, S. FORREST****, L. BRUNO**,
M.B. BITONTI**, L. SPANÒ***, D. MARIOTTI*, D. GIANNINO*

*) Institute of Biology and Agricultural Biotechnology – CNR, Section of Rome, Via Salaria km 29,300, 00016 Monterotondo Scalo, Rome

***) Dept. of Ecology, University of Calabria, Ponte Bucci, 87030 Arcavacata di Rende, Cosenza

****) Dept. of Basic and Applied Biology, University of L'Aquila, Via Vetoio, Coppito, 67010 L'Aquila

*****) Dept of Genetics and Biochemistry Clemson University, SC, USA

class I knotted-like gene, genomic organisation, expression, function by RNA interference

Knotted-like genes (*KNOX*) are homeobox containing transcription factors required to define and maintain cell identity during vegetative and reproductive patterning in the shoot apical meristem (SAM). We have further extended the characterisation of *KNOPE1*, a class I *KNOX* of peach. At the genomic level, 4 introns were identified and their position matched exactly with class I *KNATI* (*Brevipedicellus*) and *Kn1* from *A. thaliana* and *Z. mays*, respectively. Highly specific probes were used to screen a BAC library from *P. persica* cv Nemared and *KNOPE1* mapped on linkage group 7 (BAC 43A9 associated to genetic marker CC63) at approximately 23 cM. Moreover, other class I *KNOX* genes were identified on linkage group 4 and 6 using domain conserved probes.

Class I *KNOX* are known to be specifically expressed in the SAM in species with simple leaves and down regulated in leaf primordia, we previously confirmed this pattern for *KNOPE1*. However, in new experiments transcripts were detected at very low abundance in fully expanded leaves by RT-PCR and were mostly located in the vases. Transcripts were also detected in roots and forming flowers (but not in petals) suggesting a role in the development of these organs.

A virus-induced gene silencing (TRV-VIGS) approach, mediated by *A.tumefaciens*, was performed to elucidate *KNOPE1* function. The VIGS system was first tested by targeting the phytoene desaturase gene (*PDS*): mutants with chlorotic leaves were recovered and the down regulation of the endogenous *PDS* was observed. Plants injected with constructs interfering with *KNOPE1* expression exhibited reduced size, petiole shortening and deformation of leaf blade. The endogenous *KNOPE1* transcript decreased in leaves as compared to controls.