

QTLs for morphogenetic traits in *Medicago truncatula*

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Introduction Plant morphogenesis that includes growth, development and flowering date, drives a large number of agronomical important traits in both grain and forage crops. Quantitative trait locus (QTL) mapping is a way to locate zones of the genome that are involved in the variations observed in a segregating population. Co-location of QTLs and candidate genes is an indication of the involvement of the genes in the variation. The objective of this study was to analyse segregation of aerial morphogenetic traits in a mapping population of recombinant inbred lines of the model legume species *M. truncatula*, to locate QTLs and candidate genes.

Materials and methods A population of 93 recombinant inbred lines (RILs) from the F6 generation was obtained from the cross Jemalong-6 x DZA315-16. The map was built with EST-SSR markers and anonymous markers (T. Huguet, unpublished). The lines were sown four times in greenhouses in France: in 2000 at INRA, Montpellier, in 2001 at CNRS-INRA, Toulouse, in 2002 and 2003 at INRA, Lusignan. At Montpellier and Toulouse, the flowering date was individually recorded. At Lusignan, the flowering date was recorded, and transformed to degree-days. Through the growing period, the length of the first two emerging primary branches was measured 3 times a week in 2002 and twice a week in 2003. The curve of stem elongation as a function of degree-days showed a short lag phase, followed by a linear phase. It was modelled by the slope of the linear phase, named as growth rate, and the length of the stem after one week of growth in the greenhouse, named as initial length. Analyses of variance were carried out to test the effects of lines and replicates. Correlations among traits were calculated on mean values. Broad sense heritability was calculated. QTL mapping was performed using QTLCartographer with the simple interval mapping (IM) procedure. Candidate gene position was determined from data-mining of *M. truncatula* BAC sequencing or gene mapping. Co-location between QTLs and genes was analysed.

Results Large variation was recorded for all traits in each year, with several transgressive lines, when compared to the value of the parental lines. The broad sense heritabilities were high. Stem length was positively correlated to the number of internodes, the stem diameter, the initial length and the growth rate. The correlation between stem length and number of branches was low or non significant. Dry weight was correlated to stem length and diameter in both years. Leaf to stem ratio was negatively correlated to stem length, initial length and growth rate. The flowering date was strongly negatively correlated to stem length and initial length. It was not correlated to the dry weight. QTLs were found for most traits. The R² for each QTL varied from 13.0 to 62.4 %. On chromosome 7, a major QTL for the flowering date was found. This region of chromosome 7 was also involved in variation for stem length in 2002 and 2003, initial length in 2002 and 2003, number of branches in 2003, leaf to stem ratio in 2003 and stem growth rate in 2003. A gene showing homology to Constans mapped in this region. The lower part of chromosome 2 was also involved in the variation of many traits: stem length in 2002, growth rate in 2002 and 2003, dry weight in 2002, leaf to stem ratio in 2002 and 2003. A gene known to be involved in dwarfism in pea (*Le*) mapped close to this QTL. The top of chromosome 5 was implied in stem diameter in 2002 and 2003, dry weight in 2002 and 2003. Bottom of chromosome 4 was involved in the number of internodes in 2002, stem diameter, stem radius in 2002 and 2003, dry weight in 2002 and 2003. In the middle of chromosome 1, a QTL was found for the dry weight in 2002, stem radius in 2002 and 2003. No candidate gene was mapped in the region of the QTL on chromosome 1, 4 and 5.

Conclusions QTL analysis is a powerful step to identify zones of the genome involved in the variation of traits. In a single cross in *M. truncatula*, a large variation was observed for morphogenetic traits, and QTLs with a large R² were found. The current progress in genome sequencing in this species progressively offers the possibility to find candidate genes in QTL regions.