

Characterization of the *RMja* resistance gene to root-knot nematodes from the Alnem almond rootstock

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The root-knot nematodes (RKNs) *Meloidogyne* spp. may cause important damage in cultivated stone fruit orchards. In *Prunus* spp., resistance (*R*) genes to RKNs with different spectra have been identified and mapped in plum (*Ma*), peach (*RMia*) and almond (*RMja*). The two genes *Ma* and *RMia* have been identified and located on the Linkage groups 7 and 2. In order to identify the *RMja* gene of the Alnem rootstock released by Kohba and Spiegel-Roy (1976), we have created a population “Lauranne x Alnem” (LA) and we have done a fine mapping study on more 800 F2 hybrids of the LA population. Hybrids were tested in greenhouse for their resistance to *Meloidogyne javanica* and we could map the *RMja* resistance gene on the LG7 region between the two SSR markers CPPCT039 and CPPCT02. In a second step to find other polymorph molecular markers, the genome of the two parents Lauranne and Alnem were assembled by NGS sequencing and mapping on the V2 reference peach genome.

We observed that the peach gene Prupe.7G065400, annotated as the orthologous gene of *Ma* in peach, was well aligned in Alnem with a similar TNL gene which could be the *RMja* gene. As a part of the obtained Alnem sequence was missing, we were not certain there was no other candidate gene. So we constructed an Alnem BAC library in order to obtain the whole sequence of the *RMja* DNA region. With specific markers of the candidate TNL gene we screened two BAC which were sequenced using PacBio technology. The analysis of this new sequence showed that the TLN candidate gene was the sole gene that carries a complete set of canonical TNL domains. It can be considered as the *RMja* gene, although the confirmation will be effective only after functional validation by hairy roots transformation. For detecting easily this gene in Alnem hybrids obtained in our rootstock breeding program, we constructed a Kasp® marker from an Alnem SNP, specific of the resistant parent. On the 800 phenotyped hybrids, from the F2 population “Lauranne x Alnem”, all resistant hybrids were effectively predicted resistant with this Kasp® marker.