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Construction and evaluation of Near-Isogenic Lines for resistance to *Aphanomyces euteiches* in pea

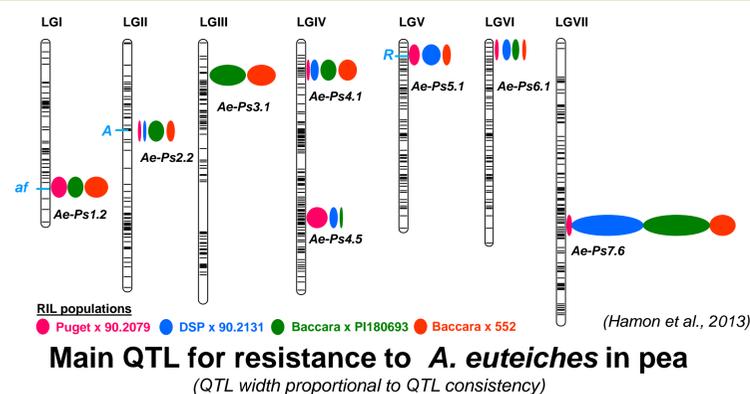
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Common root rot of pea, due to *Aphanomyces euteiches*, is one of the most damaging diseases in many pea growing countries. The development of resistant varieties is a major objective to manage the disease. Several consistent Quantitative Trait Loci (QTL) controlling partial resistance were discovered from different Recombinant Inbred Line (RIL) populations (Hamon et al., 2013).

This study aimed to validate the resistance QTL effects, individually or in combination, in different agronomic backgrounds, by creating and evaluating Near-Isogenic Lines (NILs) at 0, 1, 2 or 3 of the main resistance QTL.



NIL construction

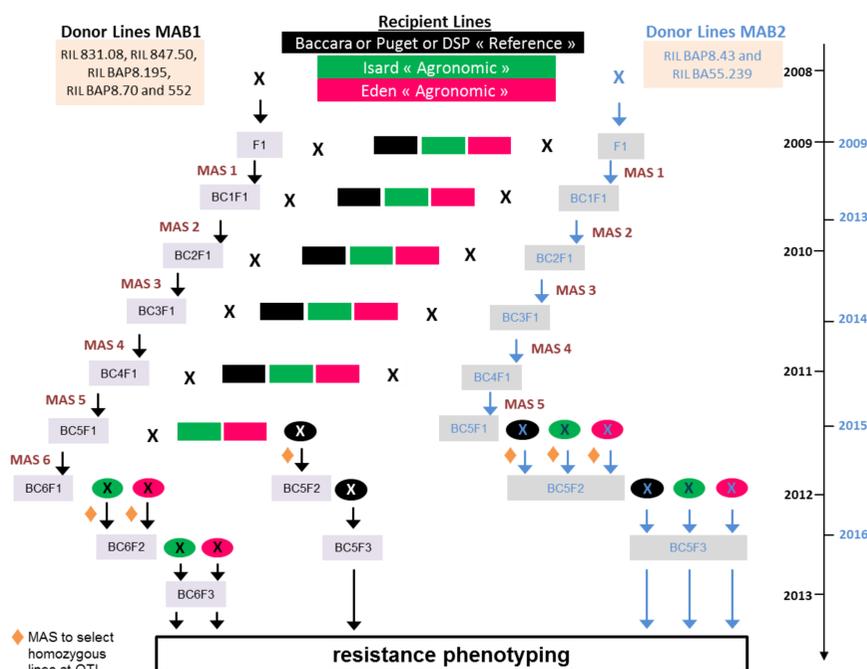


Fig. 1: Principle of Marker-Assisted Back-crossing (MAB) scheme to create NILs carrying QTL for resistance to *Aphanomyces euteiches* in pea

Between 2008 and 2016, two MAB schemes (MAB1, MAB2) were developed to create NILs derived from crosses between a total of seven donor lines (five lines in MAB1, two lines in MAB2) and three recipient lines. Resistance alleles brought by the donor lines RIL831.08, RIL847.50, BAP8.43/70/195 and BA55.239 derived from the 90.2079, 90.2131, P1180693 and 552 resistant germplasm, respectively. After five or six back-cross (BC) generations, NILs were fixed by self-pollination. Heterozygous NILs at single QTL were also selected for future fine mapping studies. SSR, eSSR and a few SNP markers of good quality (Loridon et al., 2005, Duarte et al., 2014) were used to control introgressions (MAB1/2) and return to recipient background (MAB1).

157 BC5/6 NILs from MAB1 carrying fixed resistance alleles at zero to three QTL were selected. The NILs were fingerprinted using 13,204 SNPs from the GenoPea Infinium® BeadChip (Tayeh et al., 2015). Genotypic data confirmed all the introgressions with various sizes, except at QTL *Ae-Ps3.1* in some recipient lines. The Recipient Genome Content (RGC) estimated in each NIL from the fingerprints varied from 92.5 to 100%.

36 BC5 NILs from MAB2 carrying fixed resistance alleles at zero to two QTL were selected. The NILs will be genotyped using 2,000 SNPs (Duarte et al., 2014; Boutet et al., 2016) and evaluated for resistance in controlled and field conditions.

NIL evaluation

The NILs from MAB1 were evaluated in a multi-local French field *Aphanomyces* network over two years (2014-2015) and six locations using the experimental design, as well as the root and aerial disease severity scoring scales described in Hamon et al. (2013).

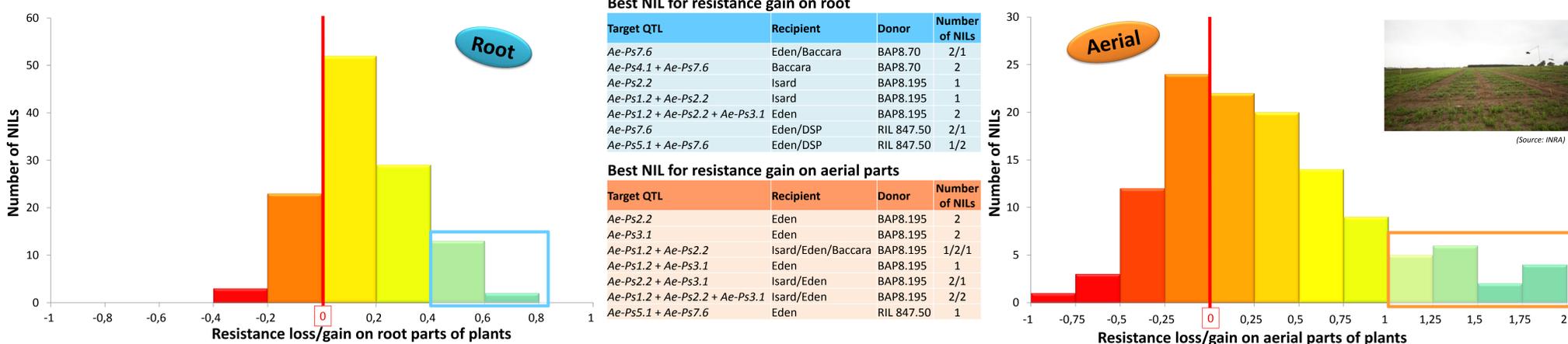


Fig. 2: Frequency distribution of resistance gain/loss provided by single or combined QTL on root and aerial parts of the plants in the MAB1 NILs
Resistance gain/loss = difference between lsmean disease severity scores on NIL without and with QTL selected from the same cross. Lsmean disease severity scores were estimated from root and aerial scoring data in 4 and 7 environments, respectively, using a linear mixed model.

Most of the NILs carrying one or several QTL showed a gain in resistance compared to NILs without a QTL.

QTL *Ae-Ps7.6*, individually or in combination with minor QTL *Ae-Ps4.1* or *Ae-Ps5.1*, increased field resistance on root parts of the plants in several recipient lines, as previously observed in controlled conditions (Lavaud et al., 2015; Fig.3).

Single or combinations of resistance alleles brought by the donor line BAP8.195 at QTL *Ae-Ps1.2*, *Ae-Ps2.2* and *Ae-Ps3.1*, increased field resistance on root and aerial parts of the plants in different genetic backgrounds. These alleles also brought undesired traits for dry pea breeding (late flowering, normal leaves, purple flowers, Hamon et al., 2013).



Fig.3: Root disease severity on NILs carrying (left) or not (right) 2 QTL

This study gives tools and information for the choice of resistance QTL useful in breeding to increase partial resistance to *A. euteiches* in future varieties.

It provides innovative plant material (NILs) for further studies about QTL mechanisms and action mode on pathogen populations and life cycle.