

Genetic and molecular bases of pea (*Pisum sativum*) resistance to pea aphid (*Acyrtosiphon pisum*)

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Social-economic context

Crops are exposed to significant yield losses due to insect pests. Therefore, large quantities of pesticides have been deployed although their use must be reduced in the coming years because of their negative impact on the environment and human health. Integrated pest management using alternative approaches, such as the development of resistant varieties to these pests, is a major challenge to maintain stable yield levels. Pea (*Pisum sativum*) is a crop that is ecologically, economically and socially valuable for its capacity to enrich the soil by fixing the atmospheric nitrogen and therefore to reduce the supply of nitrogen fertilizers. European agriculture is looking for ways to increase leguminous crops in rotations to make up for the protein deficit. However, the pea crop has been submitted to yield instability over the two last decades due to biotic stresses such as aphid infestation.

Scientific context

Acyrtosiphon pisum (the pea aphid) is a sap-sucking insect pest specialized in feeding on legume plants. This aphid species actually forms a complex of at least 15 biotypes, each one is adapted to one or a few legume species and have significantly reduced performance on other legume species (Peccoud et al., 2009). Little is known about the recognition, signalling and defense mechanisms of the plant against aphids, even less about the mechanisms involved in compatible and incompatible interactions with aphids. Several phytohormones, secondary metabolites and phloem plugging mechanisms have been revealed to be involved in plant defense against aphids, but the generalization of these results cannot be achieved for all plant-aphid interactions. Studies have shown that plant molecular defense pathways are triggered or suppressed by salivary proteins released by aphids during feeding, these proteins are thought to act in a similar way to pathogen effectors (Erb and Reymond, 2019). Hence, the molecular interactions between the specialist aphid *A. pisum* biotypes and their host and non-host legumes have to be elucidated.

Objectives

This thesis project aims to identify genetic loci and defense pathways involved in *P. sativum* resistance or susceptibility against a pea-adapted biotype and an alfalfa-adapted biotype of *A. pisum*. To that purpose, the following approaches are conducted:

(1) Identification of pea genetic loci associated with resistance against both pea aphid biotypes.

A genome wide association study (GWAS) will be conducted to identify the plant genetic loci involved in both interactions using a panel of 240 *Pisum* genotypes. In addition, resistance-associated haplotypes detected by GWAS will be validated by using recombinant inbred lines.

(2) Identification of induced defense pathways in response to aphid infestation.

Molecular pathways triggered or modulated in pea during infestation by pea and alfalfa biotypes of *A. pisum* will be identified by conducting transcriptomic, metabolomic and phytohormone experimentation on resistant and susceptible pea genotypes in infested versus non infested conditions.

(3) Functional characterization of candidate genes

A list of candidate genes and major molecular pathways involved in the compatibility and incompatibility of *P. sativum* to *A. pisum* will be established from the previous genetic and molecular studies. Reverse genetics by using mutants and virus induced gene silencing of the genes of interest will determine their functional role in these plant-aphid interactions.

Results

- GWAS was conducted using phenotypic data from the screening of the pea collection for its resistance against both *A. pisum* biotypes. Major loci that are significantly associated with each of the resistance phenotype were detected (Fig.1). In addition, for both interactions, SNPs with significant effect mapped on two common exons underlying one major locus on the chromosome 7. The results suggested that specific and common defense mechanisms are operated against adapted and non-adapted pea aphid biotypes.
- Six pea genotypes with contrasting resistant phenotype against aphids were selected to early response pathways. Significant increase of seven phytohormones were not observed after aphid infestation. RNAs extracted from pea plants challenged aphids will be sequenced and analyzed soon.

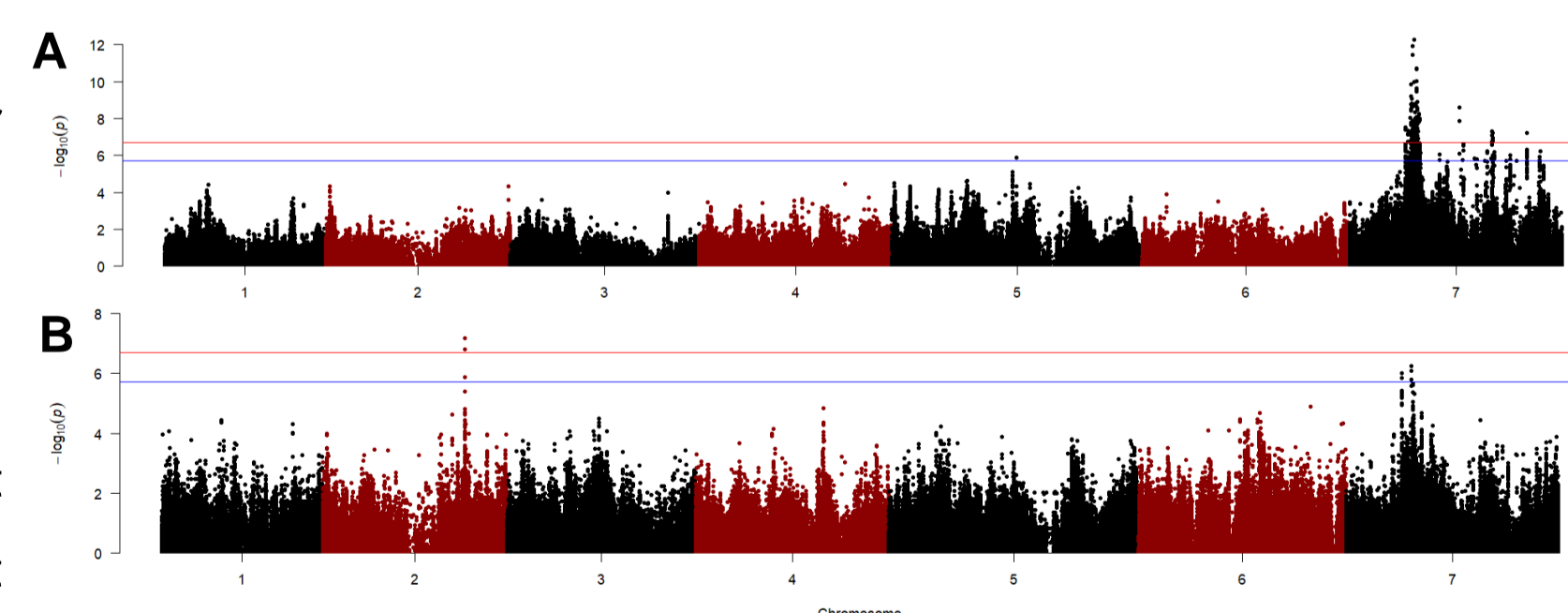


Figure 1 | Genome-wide association study (GWAS) of resistance to (A) LSR1 aphid (alfalfa biotype) and (B) ArPo28 aphid (pea biotype) in the pea collection, using 510K SNPs (MAF=5%) obtained by exome capture. X-axis corresponds to the chromosome numbers and Y-axis represents the marker-trait association p-values using the formula $-\log(pvalue)$. The horizontal red line indicates the Bonferroni correction threshold with a type 1 error rate at 10% ($P < 1.96 \times 10^{-7}$) and the horizontal blue line indicates the false discovery rate (FDR) threshold at 10% ($P < 1.96 \times 10^{-6}$).

Perspectives

Through the identification of pea markers, molecular pathways and metabolites involved in aphid resistance, this project will provide a better understanding of the whole mechanisms of recognition, signalling and defense involved in plant-aphid interaction. Such knowledge will contribute to develop resistant varieties against different aphid biotypes.

Peccoud J, Ollivier A, Plantegenest M, Simon J-C (2009) A continuum of genetic divergence from sympatric biotypes to species in the pea aphid complex.

Erb M, Reymond P (2019) Molecular Interactions Between Plants and Insect Herbivores.

