

## HEMINT

### **RNA interference for the control of hemipteran pests causing direct and indirect damages to crops: a case study to define best practices for sustainable and environment-friendly application of RNAi**

Invasive pests are a major threat to agriculture, and their impact is expected to increase with increases in world trade and ongoing climate change. Their management is largely based on chemical pesticides that have negative impacts. RNA interference (RNAi) is a sequence-specific mechanism of eukaryotes regulating gene expression and providing a defence against viral nucleic acids. Strategies based on RNAi are exploited as molecular tools for studying gene function, and are now emerging in crop protection as a very promising tool against plant pathogenic fungi, viruses and insect pests. The technology is based on the use of double-stranded RNAs (dsRNAs) to silence essential genes of a selected pest. Nevertheless, dsRNA delivery to sap-sucking insects is still an unsolved issue. In HEMINT project, RNAi will be explored for the control of two hemipteran species that cause severe damages to crops: the brown marmorated stink bug, *Halyomorpha halys* (Hemiptera: Pentatomidae), and the leafhopper *Scaphoideus titanus* (Hemiptera: Cicadellidae), main vector of Flavescence dorée (FD) phytoplasmas of grapevine. Both species currently require repeated applications of chemical insecticides for their control, thus disrupting the IPM programs developed in their absence in orchards and vineyards. Therefore, innovative and sustainable alternative strategies are urgently needed. The occurrence of RNAi in the pests under study, already reported by the Research Units involved in HEMINT, is the essential precondition for a successful outcome of the project. HEMINT is structured in 4 Work Packages (WPs), dealing with: i) coordination and dissemination activities (WP1), ii) selection of pest target genes to be silenced (WP2), iii) large scale synthesis of ds RNAs (exploiting bacteria and yeasts) that will be used in bioassays to screen target genes (WP3) and iv) optimization of delivery methods for potential field application (WP4). In particular, the pest target genes will be selected through a literature meta-analysis among those involved in lethality (*H. halys*) and vector competence (*S. titanus*), taking into account possible off-target effects on beneficial insects (pollinators, predators and parasitoids). Target genes will be initially screened on pest species, by observing the expected phenotype and by evaluating the reduction of specific transcripts. Undesirable off-target effects will be evaluated both *in silico* (by analysing sequence homology of selected genes

for several beneficials) and in vivo (by applying dsRNAs on a reasoned subset of beneficial species). Finally, in order to optimize delivery strategies, exogenous dsRNAs will be applied i) directly to the plant vascular system by petiole absorption and trunk injection ii) by leaf spraying and iii) onto *H. halys* egg masses. Exogenous application of dsRNAs is not yet approved in Europe, but the need to reduce pesticide pollution indicates that the exploration of new control tools cannot be further delayed.

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