Genetic structure and dissemination of tuf-a type stolbur phytoplasma associated with stinging nettle (*Urtica dioica* L.)

J. Johannesen¹, X. Foissac², M. Maixner³

- ¹ University of Mainz, Inst. Zoology, Saarstrasse 21, 55099 Mainz, Germany
- ² UMR 1090 INRA Université Bordeaux, France
- ³ Julius Kühn-Institut, Inst. Plant Protection in Fruit Crops and Viticulture, 54470 Bernkastel-Kues, Germany

Email: jesjo@uni-mainz.de

Stolbur 16SrXII-A phytoplasma associated with bindweed (*Convolvulus arvensis* L.) and stinging nettle (*Urtica dioica* L.) are associated with tuf-b type and tuf-a type RFLP patterns, respectively. Sequence analyses of other stolbur genes have detected further polymorphism, thus confirming a phylogenetic split between stolbur tuf-a and tuf-b types. Whereas the bindweed-associated tuf-b type is found in many other herbaceous plants, the tuf-a type is associated, as far as we know, only with stinging nettle. This species is reported to be the main host plant of the vector of stolbur in Italy and being a new host plant in Germany, where it bears the main responsibility for the increase in Bois noir disease.

We report sequence diversity of *vmp1* gene (Cimerman *et al.*, 2009) from 41 stolbur isolates (1300bp) of the tuf-a type with the aim of evaluating the geographic origin and dissemination of tuf-a type in Europe. By contrast to tuf-b type stolbur (i.e. the bindweed type), where vmp1 is phylogenetically difficult to interpret, vmp1 of tuf-a type stolbur (i.e. the stinging nettle type) is phylogenetically informative due to being more conserved. Vmp1 of the tuf-a type strain was detected for a penta-peptide sequence "Asp-Val-Ala-Asn-Asn", thus confirming the uniqueness of the tuf-a type strain. We found 14 genotypes with maximum 2 % sequence divergence. Diversity in Germany and Switzerland was extremely low. Nineteen sequences out of 20 were identical (average nucleotide diversity over loci = 0.000230). Ten isolates analysed from Northern and Central Italy (including one from South-Eastern France) all showed different genotypes and were very diverse (average nucleotide diversity = 0.010729). We observed three genotypes among 11 Slovenian and Croatian isolates (average nucleotide diversity = 0.000780). The most common genotype was found in seven isolates; this genotype was also found once in Italy. As high genetic diversity is often associated with centres of origins, Italy may constitute the geographic origin of tuf-a type stolbur phytoplasmas. Genetic diversity decreased away from Italy, indicating a longer evolution in Italy on stinging nettle, and bottleneck effects in more recently colonised areas. Phylogenetic analysis of the nettle-stolbur genotypes suggests that nettle-stolbur arrived in Germany via Italy or France and not via Slovenia, as indicated for the vector *Hyalesthes obsoletus* Signoret (Johannesen *et al.*, 2008). The lack of monophyly of Slovenian/Croatian and German genotypes coupled with the observation of genetic bottlenecks in both areas but for different genotypes, suggests that Slovenia/Croatia was not the source area of German nettle-stolbur. If our hypothesis is correct, it implies that stolbur tuf-a type and the vector *H. obsoletus* have experienced independent dispersal dynamics in Europe. This hypothesis is presently being tested.

Keywords: VMP1, host-plant strains, dispersal.

References

Cimerman A., Pacifico D., Salar P., Marzachì C., Foissac X., 2009. Striking diversity of *vmp1*, a variable gene encoding a putative membrane protein of the stolbur phytoplasma. Applied and Environmental Microbiology 75, 2951-2957.

Johannesen J., Lux B., Michel K., Seitz A., Maixner M., 2008. Invasion biology and host specificity of the grapevine yellows disease vector *Hyalesthes obsoletus* in Europe. Entomologia Experimentalis et Applicata 126, 217-227.