

## Occurrence and distribution of *Scaphoideus titanus* in multiple outbreaks of “flavescence dorée” in Serbia

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### Abstract

A total of 23 viticultural regions in Serbia were surveyed from 2004 to 2006 to determine the occurrence and distribution of *Scaphoideus titanus*, vector of grapevine phytoplasma “flavescence dorée”. Grapevine samples from over 600 plants showing suspected symptoms of phytoplasma infection, were collected from all surveyed regions for subsequent molecular identification and determination of FD incidence. Presence of *S. titanus* was surveyed in all areas and collected samples were analyzed with PCR/RFLP for FD presence confirmation. With exception of southern region of Pčinja, *S. titanus* was detected in all surveyed regions, including areas close to Croatian, Hungarian, Romanian and Bulgarian state borders. Significant populations of this leafhopper were recorded in vineyards, as well as, on wild *Vitis* plants, showing wide distribution of this species in Serbia. FD phytoplasma was registered both, in vector and in grapevine in 8 regions and its presence was correlated with high density of *S. titanus* populations in surveyed vineyards. Incidence of symptomatic plants in vineyards ranged from 10 to 100% in most endangered regions, while infestation level of *S. titanus* with FD ranged from 12.5 to 45%.

**Key words:** *Scaphoideus titanus*, “flavescence dorée”, grapevine, epidemiology, distribution.

### Introduction

“Flavescence dorée” (FD) is a serious and severe disease of grapevine (*Vitis vinifera* L.) in temperate areas of several European countries (France, Italy, Spain and Switzerland). The disease is transmitted by *Scaphoideus titanus* Ball (Auchenorrhyncha Cicadellidae), a leafhopper of North American origin that became the most serious threat for dispersal of FD in Europe. Presence of *S. titanus* in Serbian vineyards was reported with considerable delay (Magud and Toševski, 2004), shortly after first report of FD infected vineyards (Duduk *et al.*, 2003). Župa Aleksandrovac and Niš, two distinct regions in south central Serbia, were the only regions with confirmed presence of FD (Duduk *et al.*, 2004; Kuzmanović *et al.*, 2004).

The aims of this study were: i) to determine the occurrence and distribution of *S. titanus* in Serbia, ii) to verify distribution of FD phytoplasma in Serbia.

### Materials and methods

Insects sampling was carried out between 2004-2006 by sweeping with entomological net and using yellow sticky traps to determine presence and abundance of *S. titanus*. Visual inspection of vineyards was performed at the beginning of nymph emergence, from mid May to mid June. Sweeping was carried out from mid June to the end of September. During that period, yellow sticky traps were placed at 5 plots (10x10 m) per site, in at least 5 selected vineyards, following geographic wide-spread of surveyed vineyards regions. Yellow sticky traps were also randomly exposed on wild growing *Vitis*

spp. populations between neighboring vineyards. According to the number of *S. titanus* captured on yellow sticky traps every 15 days (during July and August), 1-25 collected specimens were considered as low population density, 25-100 as moderate and over 100 as high population density. Highest number of *S. titanus* specimens detected in surveyed vineyards/sites was accepted for final interpretation of results.

Insect material collected by sweeping was maintained in 80% alcohol for later determination and phytoplasma identification. Single specimens of *S. titanus* were PCR analyzed for presence of FD to estimate infection rate on sites where FD was identified in plant material, or in pools of 5-10 specimens in regions where FD was detected.

Plant material showing symptoms typical for phytoplasma infection was collected from beginning of July to the end of September during 2004-2006. Incidence of phytoplasma diseased grapevines was presented as percent of plants with symptoms in vineyards. Infected plants were collected and labeled for PCR analyses to confirm presence and type of phytoplasma. Depending on the size of vineyards and incidence of diseased vines, samples were collected from 1 to 10 plants per site.

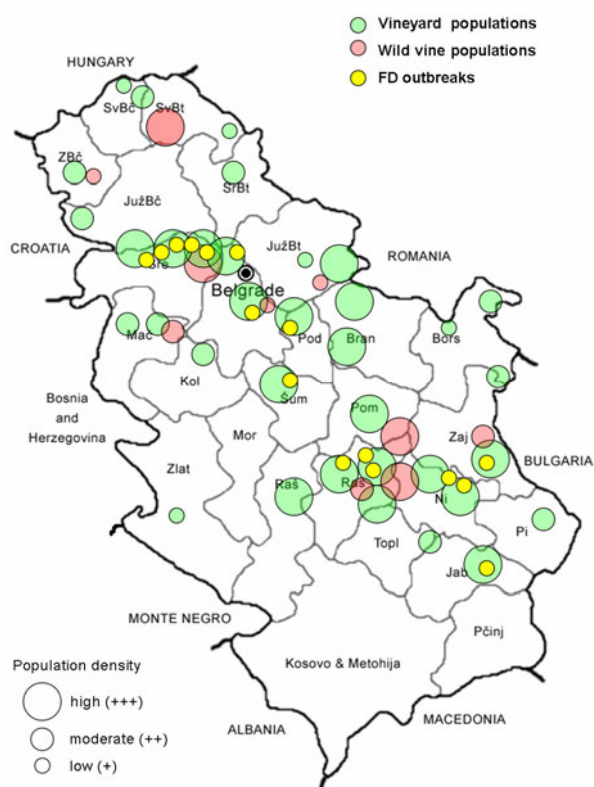
DNA extracted from plant and insect material was amplified by nested PCR according to previously reported protocols (Angelini *et al.*, 2001; Martini *et al.*, 2002). RFLP analyses were performed to determine FD phytoplasma ribosomal subgroup by overnight digestion of nested PCR product with *TruI* and *TaqI* enzymes.

A total of 163 vineyards were inspected during this study and over 600 symptomatic plants and 225 *S. titanus* specimens were analyzed for phytoplasma presence.

## Results

Presence of *S. titanus* was determined in every traditional vine growing region of Serbia, with exception of southern region of Pčinja. Territory of Kosovo was excluded from the survey. Population size of this leafhopper was fluctuating from low to extremely high, depending on the extent of applied chemical measures. Obtained results about wide distribution and dense populations indicate that *S. titanus* had been present in vineyards, as well as, in wild *Vitis* plants in Serbia, for a long period of time. Presence of this leafhopper was also verified in the border zone with Croatia, Bosnia and Herzegovina, Hungary, Romania and Bulgaria.

PCR and RFLP results confirmed presence of FD-C phytoplasma in grapevine and *S. titanus* in 8 major vine growing areas of Serbia (figure 1). Incidence of symptomatic plants in these vineyards ranged from 10 to 100% in most endangered regions, while infestation level of *S. titanus* with FD ranged from 12.5 to 45%.



**Figure 1.** Distribution and population density of *S. titanus* and occurrence of FD phytoplasma in Serbia. (In colour at [www.bulletinofinsectology.org](http://www.bulletinofinsectology.org)).

## Discussion

Data presented here are results of three years survey performed in Serbia, studying distribution and dispersal of *S. titanus*. These results show wide distribution of *S. titanus* in Serbia. High density of vector's populations is correlated with multiple outbreaks of FD in at least 8 distinct regions, which covers about 39.6% of vine production. Presence of *S. titanus* close to borders of the neighboring countries could have a negative influence on phytosanitary situation related with potential spread of *S. titanus* and FD in southeastern Europe.

Phytosanitary situation in vineyards of Serbia alerts urgent control of this quarantine disease by conducting phytosanitary inspection of vineyards, eradication of infected grapevines, destroying natural reservoirs of phytoplasma, as well as, monitoring and control of vector.

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