

## Grapevine yellows survey in grape growing areas of four districts in Bologna province

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### Abstract

Samples of symptomatic grapevines were collected from 2002 to 2006 in different localities from the main grapevine growing areas of Bologna province (Emilia-Romagna region). Total nucleic acids were extracted from leaf midribs following molecular protocols. "Flavescence dorée" and "bois noir" were detected. In four districts was carried out more detailed inspections.

**Key words:** Grapevine yellows, monitoring, "flavescence dorée", "bois noir", molecular detection.

### Introduction

Grapevine yellows are diseases associated to phytoplasmas present in all grape growing areas of Emilia-Romagna region and they are increasing. An important outbreak of grapevine yellows was observed during 1999-2001 and "flavescence dorée" (FD) and "bois noir" (BN) phytoplasmas were detected by molecular analysis of symptomatic samples collected in surveyed vineyards in western provinces while only BN was found in central-eastern provinces.

During 2002 FD was detected for the first time also in western area of Bologna province (in the flat land) considered free from the disease until this date, although the vectors *Scaphoideus titanus* Ball was reported for many years.

In Italy is in force a ministerial decree of compulsory phytosanitary measures with the aim to eradicate plants infected by FD.

In order to verify the incidence of yellows diseases, more detailed survey started in all the vineyards of four important grapevine districts (Argelato, Calderara di Reno, Sala Bolognese, San Giovanni in Persiceto) of this province.

### Materials and methods

During inspections carried out from 2002 to 2006, 388 grapevine symptomatic samples were collected in different grapevine growing areas of Bologna province. In 2006, during summer and autumn, visual checks were carried out in all the 463 vineyards (467 hectares) present in four grapevine districts and the percentage of symptomatic plants was calculated referred to each investigated district.

To ascertain the presence and identify of phytoplasmas, total DNAs were assayed by PCR on ribosomal sequence using universal primers P1/P7 (Deng and Hiruki, 1991; Smart *et al.*, 1996) followed by nested PCR with specific primers R16(V)F1/R1 (Lee *et al.*, 1994) for FD detection, and fStol/rStol (Maixner *et al.*, 1995)

for BN detection. As regards of FD strains characterization, positive samples were tested in nested-PCR using non-ribosomal primers specific for group V, FD9f2/r followed from FD9f3/r2 (Angelini *et al.*, 2001). The amplified PCR product was further RFLP analyzed by digestion with *TaqI* restriction enzyme.

### Results

A high percentage of the 388 symptomatic grapevine samples collected in Bologna province result infected by phytoplasmas in nested-specific PCR: 56 samples (15%) were infected by FD phytoplasmas, all resulted, after characterization, belonging to subgroup 16SrV-D except one sample infected by phytoplasmas of the subgroup 16SrV-C; 265 samples (68%) were infected by BN, 9 samples (2%) were infected by both phytoplasmas and 58 samples (15%) resulted negative. The grapevine varieties infected were: Albana, Barbera, Cabernet Sauvignon, Chardonnay, Grechetto, Lambrusco Grasparossa, Lambrusco Salamino, Montuni, Moscato, Pignoletto, Raboso, Sangiovese e Trebbiano Romagnolo.

In the four districts monitored, a high percentage (66.74%) of vineyards showed grapevine yellows symptoms with different incidence. The percentage of vineyards with symptomatic plants was: 92.77% (77 vineyards infected /83 vineyards surveyed) in Argelato district, 56.25% (45 vineyards infected/80 vineyards surveyed) in Calderara di Reno district, 80.25% (65 vineyards infected/81 vineyards surveyed) in Sala Bolognese district and 55.71% (122 vineyards infected/219 vineyards surveyed) in San Giovanni in Persiceto district.

Despite of the high number of vineyards with symptomatic plants, only a low percentage (2.2%) of plants showed yellows symptoms in four districts, with the following distribution: 8.0% (4,884/60,806) in Argelato district, 1.5% (1,733/11,8437) in Calderara di Reno district, 2.3% (5,144/22,1526) in Sala Bolognese district and 0.9% (1,970/21,6749) in San Giovanni in Persiceto district.

## Discussion

Grapevine yellows symptoms were widespread in Bologna province even if they appeared in most vineyards only on a few plants. Molecular analysis confirmed the presence of phytoplasmas in high percentage of sample tested (85%): BN (68%) was widespread in all the vineyards while FD (15%) was mainly located in the western parts of grape growing areas. The detection of FD infected samples (15% only FD and 2% mixed infection with BN), in vineyards where *S. titanus* occurs, represents a phytosanitary risk: measures to contain FD were and are applied by uprooting infected plants and containing *S. titanus* presence by using appropriate insecticides.

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