

Almond witches'-broom phytoplasma (*Candidatus Phytoplasma phoenicium*): a real threat to almond, peach and nectarine.

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Introduction

Within less than a decade, Almond witches'-broom (AlmWB) phytoplasma killed over a hundred thousand almond trees in Lebanon (Abou-Jawdah et al., 2002). AlmWB belongs to the pigeon pea witches' broom group (16SrIX), and the scientific name (*Candidatus Phytoplasma phoenicium*) was suggested (Verdin et al., 2003). Grafting experiments revealed that AlmWB may also affect peaches and nectarines (Abou-Jawdah et al., 2003). Later on, a similar disease was reported in Iran (Verdin et al., 2003; Salehi et al., 2006). This disease is still spreading on almond trees to new areas in North Lebanon, but more recently shoot proliferation with succulent small light green leaves were observed on peach and nectarine in South Lebanon, where the disease seemed to be spreading relatively fast. DNA sequencing showed over 99% sequence homology with AlmWB (Abou-Jawdah et al. 2008). This report shows that epidemics of AlmWB may occur also on peach and nectarine under field conditions, and strongly suggests the presence of an efficient vector.

Materials and methods

Surveys on major stone fruit production areas in South Lebanon and in West Bekaa for the presence of proliferation symptoms were conducted in 2008 and 2009. Leaf samples were collected from plants showing proliferation symptoms. DNA extraction from leaf midribs was performed using the modified CTAB protocol as previously described (Zhang et al., 1998; Abou-Jawdah et al., 2002). Samples from different locations were analysed by nested PCR using universal primers (P1/P7 and nested PCR R16F2/R2) as described previously (Gunderson et al. 1996). The amplicons were sequenced and the resulting sequences were subjected to Blast analysis.

Results

In 2008, About 110 nectarine and peach trees in two orchards in the Sarada plain (South Lebanon) showed characteristic symptoms of shoot proliferation; smaller leaves with a light green color (Fig. 1). Nested PCR and sequencing confirmed infection by AlmWB. Farmers eradicated all symptomatic trees.



Fig. 1 Characteristic symptoms of AlmWB on peach and nectarine. A. Shoot proliferation on nectarine, B. Comparison of infected (red arrow) vs. healthy peach tree, C. Close up view: infected nectarine branch (left) vs. healthy one (right)

In 2009, the survey conducted in South Lebanon and West Bekaa showed that only a limited number of peach and nectarine trees are symptomatic; however, they were spread over a large area (Fig. 2).

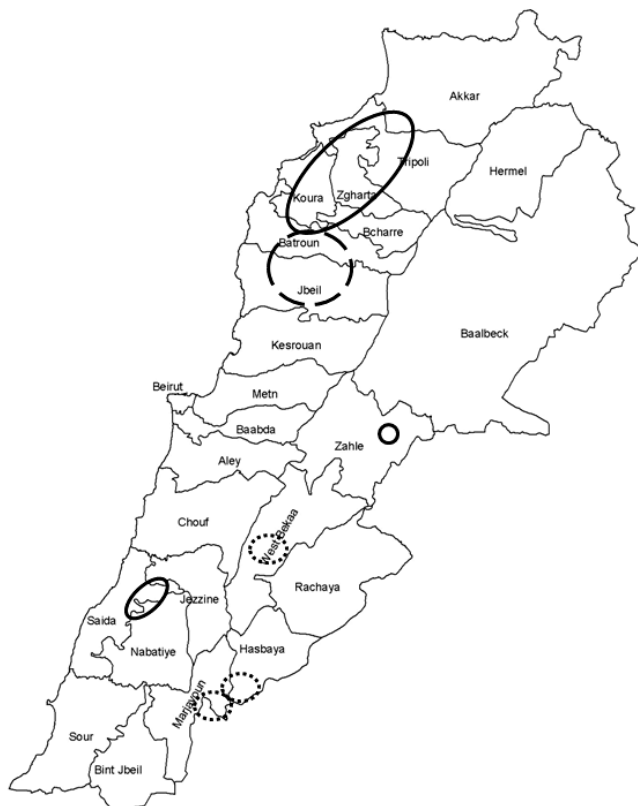


Fig. 2 Map of the AlmWB spread, starting in 2001 (full circles) and surveyed in 2009 (dotted circles); dashed circles show the area of a rapid spread that has not been yet precisely surveyed.

The evolution of symptoms during the season was monitored, showing an earlier flowering and development (Fig. 3A) and later on characteristic shoot proliferation (Fig. 1) and in rare cases witches'-broom symptoms (Fig. 3D). Most infected trees did not set any fruits, but some trees bore a limited number of deformed fruits (Fig. 3C) and abnormal flowers (phyllody) (Fig. 3B). Blast analysis of the 16S rDNA sequences obtained from 15 samples collected from several locations (South and Bekaa) revealed over 99% sequence homology with AlmWB.

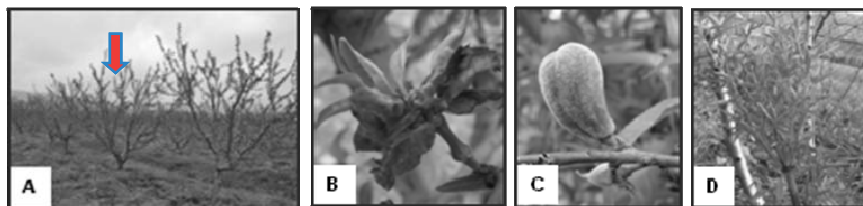


Fig. 3 Characteristic symptoms of AlmWB on peach and nectarine. A. Early in the season, infected nectarine tree (red arrow) between healthy ones, B. Phyllody or transformation of flowers into leaf and stem structures of peach, C. Deformed peach fruit, D. Witches'-broom on peach.

In the North the disease on almond is progressing further towards central Lebanon, and has reached the Koura and Jbeil districts, but no surveys have been undertaken yet to monitor precisely the infected locations.

Discussion

AlmWB is a real threat to almond, peach and nectarine, not only in Lebanon, but also worldwide since the disease spreads from sea level to an elevation of about 1000 m. Furthermore, its natural spread on certified imported seedlings, clearly indicates the presence of an efficient vector(s). Further studies are needed on the epidemiology of the disease, the vector(s) involved, alternative hosts, varietal sensitivity/resistance, to decide if almond, peach and nectarine are a dead end host for AlmWB or if they constitute a good source of inoculum. Regional and international cooperation should be initiated and effective legislation and control measures should be implemented to prevent the further spread of AlmWB, which may prove to be more devastating to stone fruit production than *Plum pox virus*.

Aknowledgements

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Literature

- Abou-Jawdah, Y.; Dakhil, H.; El-Mehtar, S.; Lee, I.M.; 2003: Almond witches'-broom phytoplasma, a potential threat to almond, peach and nectarine. *Canadian Journal of Plant Pathology* **25**, 28–32.
- Abou-Jawdah, Y.; Karakashian, A.; Sobh, H.; Martini, M.; Lee, I.M.; 2002: An epidemic of almond witches'-broom in Lebanon: classification and phylogenetic relationship of the associated phytoplasma. *Plant Disease* **86**, 477–484.
- Abou-Jawdah, Y.; Sobh, H.; Akkary, M.; 2009: First report of Almond witches' broom phytoplasma ('Candidatus *Phytoplasma phoenicium*') causing a severe disease on nectarine and peach trees in Lebanon. *OEPP/EPPO Bulletin* **39**, 94–98.
- Gundersen, D.E.; Lee, I.M.; 1996: Ultrasensitive detection of phytoplasmas by nested-PCR assays using two universal primer pairs. *Phytopathol. Mediterr.* **35**:144-151
- Salehi, M.; Izadpanah, K.; Heydarnejad, J.; 2006: Characterization of a new almond witches' broom phytoplasma in Iran. *Journal of Phytopathology* **154**, 386–391.
- Verdin, E.; Salar, P.; Danet, J.L.; Choueiri, E.; Jreijiri, F.; El Zammar, S.; 2003: 'Candidatus *Phytoplasma phoenicium*' sp. nov. a new phytoplasma associated with an emerging lethal disease of almond trees in Lebanon and Iran. *International Journal of Systematic Bacteriology* **53**, 833–838.
- Zhang, Y.P.; Uyemoto, J.K.; Kirkpatrick, B.C.; 1998: A small scale procedure for extracting nucleic acids from woody plants infected with various phytopathogens for PCR assay. *J. Virol. Methods.* **71** (1):45-50.