Etiology of phytoplasma-associated diseases of almond, pomegranate, and grapevine in Jordan

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In this study, a national survey on phytoplasma-associated diseases was conducted in Jordan from 2019 to 2021 targeting almond, pomegranate, and grapevine, three of the main crops cultivated in all country as commercials and family farming. The activities included: (i) monitoring and sampling symptomatic and symptomless plants from early summer to autumn; (ii) total nucleic acids extraction and phytoplama detection by 16S rDNA amplification in nested PCRs using the primer pairs P1/P7 followed by F1/R0; (iii) sequencing and bioinformatic analyses (BlastN, iPhyClassifier) of F1/R0 amplicons. During field surveys, almond yellows and witches'-broom (incidence around 40%), pomegranate exhibiting leaf chromatic alteration and rolling, little leaf and witches'-broom (incidence around 70%), and grapevine yellows (incidence around 10%) were observed. Molecular detection and 16S rDNA nucleotide sequence analyses revealed the presence of different 'Candidatus Phytoplasma' species within samples from symptomatic plants, while no amplification was obtained from symptomless plant samples. 'Ca. P. solani'-related strains was identified in all almond trees showing yellowing, while seven phytoplasma species were found in almond witches'-broom affected plants, including 'Ca. P. ulmi'-related strains (identified in 28% of analyzed trees), 'Ca. P. omanense'-related strains (24%), 'Ca. P. asteris'-related strains (12%), 'Ca. P. aurantifolia'-related strains (12%), 'Ca. P. pyri'related strains (12%), 'Ca. P. trifolii'-related strains (6%), and 'Ca. P. phoenicium'-related strains (6%). In pomegranate symptomatic plants, four different phytoplasma species were identified: 'Ca. P. solani'-related strains (identified in 52% of analyzed trees), 'Ca. P. aurantifolia'-related strains (21%), 'Ca. P. ulmi'-related strains (16%), and 'Ca. P. asteris'related strains (11%). Interestingly, 'Ca. P. ulmi', Ca. P. pyri', and 'Ca. P. omanense' in association with almond, and 'Ca. P. ulmi' in association with pomegranate are reported for the first time in this study. The other phytoplasma species identified in almond and pomegranate were previously reported in the Middle East. In grapevine yellows affected plants, three phytoplasma species were found: 'Ca. P. solani'-related strains (identified in 70% of analyzed plants), 'Ca. P. omanense'-related strains (25%), and 'Ca. P. aurantifolia'related strains (5%). Such phytoplasmas were largely reported in previous works in association with grapevine yellows in the Middle East. Data obtained in this study revealed a great genetic diversity of phytoplasmas infecting important crops in Jordan. Further studies concerning the epidemiology of these phytoplasma-associated diseases, including the identification of putative insect vectors and reservoir plants, are in progress. Overall results will allow developing integrated strategies for the management of such diseases.