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NEW DISEASE REPORT

A new disease of spring onion associated with a '*Candidatus* Phytoplasma asteris'-related strain in Turkey

I. Erguven | **M. E. Guldur** | **H. Akkurak** | **E. Simsek** | **H. Ayvaci** | **M. Dikilitas**

Faculty of Agriculture, Department of Plant Protection, Harran University, Şanlıurfa, Turkey

Correspondence
H. Akkurak, Faculty of Agriculture, Department of Plant Protection, Harran University, 63290, Şanlıurfa, Turkey
E-mail: gumush199@gmail.com

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Spring onion (*Allium fistulosum*) is an economically important crop and has a significant place in vegetable production in Turkey with an annual production of 2 million tonnes. They are grown throughout the year around the city of Adana (Adana province) which provides a 6.59 (4.24 tones/da) of the country's production. In 2019, leaves of field-grown onions, grown for commercial sale, exhibited typical phytoplasma symptoms such as yellowing, distortion, deformation and stunting (Figure 1).

Samples of fresh leaves were obtained from onion plants exhibiting yellowing, distortion and stunting (n:16) or from asymptomatic plants (n:8), and DNA was extracted using a CTAB method (Doyle & Doyle 1987). Template DNA was tested with a nested PCR using R16F1/RO primers in the first round PCR followed by R16F2n/R2 primers in the nested round to amplify a 1,250 bp DNA fragment from the 16S rRNA gene (Duduk et al., 2013). Amplicons of the expected size were obtained from all diseased samples but not from

FIGURE 1 Symptoms on onion plants collected from the field in Adana, Turkey and infected with phytoplasma: a) twisted leaf (white arrow), b) deformation and distortion, c) yellowing and stunting (left) and healthy plant (right)

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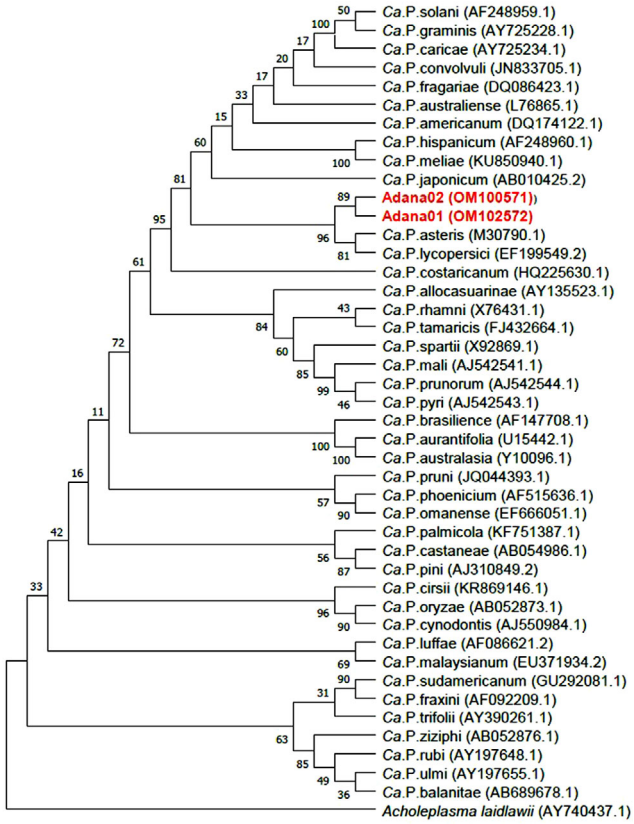

FIGURE 2 Phylogenetic tree constructed by the neighbour-joining method comparing partial 16S rDNA sequence (1,200 kb) of isolates Adana01 and Adana02 (in red) with 42 other '*Ca.* Phytoplasma species'. Numbers on branches are bootstrap values of 1000 replicates and the bar indicates the number of substitutions per nucleotide position. *Acholeplasma laidlawii* was used as the outgroup

the symptomless plants. The resulting amplicons were purified using ExoSAP-IT™ reagent (ThermoFisher Scientific, USA), and sequenced in both directions. The 16SrDNA partial sequences were assembled using the MEGAX software programme and compared using BLASTn analysis. The 16SrDNA partial sequences from all 16 diseased plants were identical and two representative isolates (Adana01 and Adana02; GenBank Accession Nos. OM100571 and OM102572) showed 99.8% identity with the 16SrI-B phytoplasma subgroup. A phylogenetic tree was constructed by the neighbour-joining method using the MEGAX software (Figure 2). The isolates shared 99.8% identity with that of the '*Ca* Phytoplasma. asteris' reference strain (M30790).

Previously, Khadharir et al. (2002) stated that severe leaf yellowing, reduced yield and stunting were evident in phytoplasma-infected onions in Canada. Phytoplasmas collected from these onions belonged to the 16SrI-A subgroup. On the other hand, Cho et al. (2019) reported that the phytoplasma agent causing yellowing and stunting in onions was associated with the 16SrI-B subgroup of '*Candidatus* Phytoplasma asteris' in Taiwan. Recently, Akkurak et al. (2021) reported that a phytoplasma causing yellowing symptoms in lettuce in the Turkish province of Sanliurfa belonged to the 16SrI-B subgroup. Caglayan et al. (2013) reported that a phytoplasma causing sweet cherry in Turkey belonged to the 16SrI-B subgroup. To the best of our knowledge, this is a new disease of '*Ca.* Phytoplasma asteris'-related strain (16SrI-B subgroup) causing significant crop reductions (1.3%) in onion plants in Turkey. The disease is exhibiting an increased trend in terms of prevalance and frequency as the symptoms of disease infected almost 20% of the plants in fields and gardens in a year period.

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ORCID

M. E. Guldur https://orcid.org/0000-0002-3374-5602
H. Akkurak https://orcid.org/0000-0003-1196-5230
E. Simsek https://orcid.org/0000-0003-4984-4223
H. Ayvaci https://orcid.org/0000-0002-5620-4147
M. Dikilitas https://orcid.org/0000-0002-7399-4750

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