



First report of '*Candidatus Phytoplasma aurantifolia*' (16SrII) associated with potato purple top in San Gabriel-Carchi, Ecuador

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Received: 24 Jul 2015. Published: 15 Nov 2015. Keywords: nested PCR, PPT phytoplasma, 16S rRNA

During April 2015 common phytoplasma symptoms such as potato purple top, aerial tubers and witches' broom were observed in 70% of commercial potato plantations (*Solanum tuberosum*) in San Gabriel-Carchi, Ecuador (Fig. 1). In Ecuador, there had not been any previous reports of this disease. However in Carchi the disease is rapidly spreading and is causing serious economic losses to potato producers. To confirm the association of symptoms with a phytoplasma infection, total DNA was extracted from 300 mg of leaf midribs and petioles from symptom-bearing (five samples) and symptomless (five samples) potato plants using a CTAB method (Doyle & Doyle, 1990) and was purified with PureLink[®] Plant Total DNA Purification Kit (Invitrogen™, ThermoFisher Scientific). Phytoplasma DNA was amplified by nested PCR with the universal primers P1/P7 (Deng & Hiruki, 1991; Schneider *et al.*, 1995) followed by fU5/rU3 (Seemüller *et al.*, 1994). Nested PCR produced amplicons corresponding to the phytoplasma 16S ribosomal DNA of 0.8 kb in two samples collected from diseased plants but no products in healthy plants. The nested PCR products were purified and sequenced using commercial facilities (Macrogen, MD, USA). The sequences of our samples were deposited in NCBI with GenBank Accession Nos. KT312845 and KT312846. BLAST analysis showed the highest sequence identity (99%) with phytoplasmas belonging to group 16SrII '*Candidatus Phytoplasma aurantifolia*' (formerly called peanut witches' broom phytoplasma), including orbea gigantea phytoplasma (HG421073), brinjal little leaf phytoplasma (KP866409), sesame phyllody phytoplasma (KP297862) and tomato big bud (JQ868448) among others.

Phylogenetic analysis (Fig. 2) using MEGA version 6.0 (Tamura *et al.*, 2013) constructed using partial sequences of 16S rRNA gene (0.8 kb) confirmed that the two sequences (potato purple top-Mv1 and potato purple top-U1) were clustered with other members of the 16SrII group. Our findings agree with Santos *et al.* (2010) who reported peanut witches'-broom group (16SrII) '*Candidatus Phytoplasma aurantifolia*' as the causal agent of potato purple top in Guanajuato and Sinaloa, México. To our knowledge we report for the first time in Ecuador the association of the '*Ca. P. aurantifolia*' (16SrII) group with symptoms characteristic of purple top, aerial tubers and witches' broom in potato plantations. These findings deserve further investigation, for example to complete the molecular

characterisation of this phytoplasma using other taxonomically relevant genes and to describe its association with potential vectors in the field.

Acknowledgements

We would like to thank Agronomist Julio Cañar (Farmagro-Ecuador) for his technical assistance in the field sample collection. The authors appreciate the collaboration of Dr. Antonio León (Universidad San Francisco de Quito-Laboratorio de Biotecnología Agrícola y de Alimentos) for allowing the use of specialised lab equipment.

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Figure 1

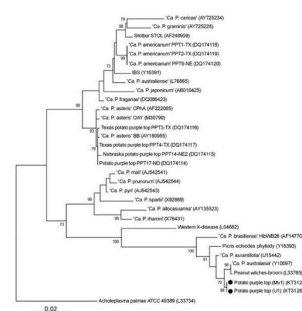


Figure 2

To cite this report: Caicedo J, Crizón M, Pozo A, Cevallos A, Simbaña L, Rivera L, Arahana V, 2015. First report of '*Candidatus Phytoplasma aurantifolia*' (16SrII) associated with potato purple top in San Gabriel-Carchi, Ecuador. *New Disease Reports* **32**, 20.

<http://dx.doi.org/10.5197/j.2044-0588.2015.032.020>

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