Diseases Caused by Bacteria and Phytoplasmas

First Report of a ‘*Candidatus Phytoplasma pruni*’-Related Strain Associated with Commercial Poinsettias in Canada

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The poinsettia, *Euphorbia pulcherrima* Willd. ex Klotzsch (family Euphorbiaceae) is the second-largest potted ornamental crop in Canada; approximately 4.2 million pots per year are grown in Ontario greenhouses (Agriculture and Agri-Food Canada 2020). Their commercially popular compact appearance is due to a branch-inducing factor, the PoiBI phytoplasma (Lee et al. 1995). Millions of potted poinsettias are the most sold Christmas decoration around the globe, including Canada. However, the Canadian poinsettia phytoplasma strain has not yet been characterized. Potted poinsettias exhibiting smaller size bracts than normal and corrugated leaves were observed in December 2019 in a flower shop in Essex County, Ontario. Commercial cultivars of *E. pulcherrima* often carry *Poinsettia latent virus* (PnLV, formerly *Poinsettia cryptic virus*) without inducing symptoms (aus dem Siepen et al. 2005) and poinsettia mosaic virus (PnMV), causing unapparent to light mottling symptoms. Because the symptoms observed in the Ontario poinsettias were not indicative of a possible PnMV or PnLV infection, and considering that reduction of leaf size is a typical phytoplasma-induced symptom, only phytoplasmas were investigated. Nested PCRs with primers amplifying the phytoplasma 16S rRNA (Gundersen and Lee 1996) and elongation factor Tu (*tuf*) (Makarova et al. 2012) genes were performed on DNA extracts (FAST DNA Spin Kit, MP Biomedicals, U.S.A.) of five symptomatic plants. Total DNA from healthy garden poinsettia plants was used as a negative control. R16F2n/R2 and *tuf* PCR amplicons, obtained from symptomatic poinsettias only, were purified and sequenced. BLAST comparison (<https://blast.ncbi.nlm.nih.gov>) of the R16F2n/R2 (GenBank accession no. MW363494) and *tuf* (GenBank accession no. MW590959) sequences of the Ontario poinsettia phytoplasma yielded 99% sequence identity with those of

phytoplasmas of group 16SrIII ‘*Candidatus Phytoplasma pruni*’. The in silico restriction fragment length polymorphism patterns from *iPhyClassifier* (<https://plantpathology.ba.ars.usda.gov/cgi-bin/resource/iphyclassifier.cgi>) were identical to that of the *Prunus persica* phytoplasma (GenBank accession no. JQ044393), subgroup 16SrIII-A, with a similarity coefficient of 0.99. Results differed from those in which the PoiBI phytoplasma was identified as a member of subgroup 16SrIII-H, including the United States (Lee et al. 1998), Italy (Pondrelli et al. 2002), and Korea (Chung and Choi 2010). Phylogenetic trees constructed with the neighbor-joining method (MEGA 7.0) based on the 16S rRNA and the *tuf* gene sequences placed the Ontario poinsettia phytoplasma within the 16SrIII phytoplasma clade, closely related to the subgroup 16SrIII-A. In Canada, the 16SrIII phytoplasma group has been associated with peach-X disease and reported in cherry, clover, and grapevine (Olivier et al. 2009), and more recently in *Trillium* sp. (Arocha-Rosete et al. 2016). Further studies will aid in determining whether or not mixed infections with other phytoplasma groups can occur, including 16SrI (‘*Ca. P. asteris*’) or 16SrXII (‘*Ca. P. solani*’), as reported in 16SrIII-H phytoplasma infected poinsettias (Pondrelli et al. 2002). This is the first report of the occurrence of a 16SrIII-A phytoplasma strain in *E. pulcherrima* in Canada in commercial poinsettias. Because 16SrIII-A phytoplasma is an EPPO A1 quarantine pest, management of infected poinsettias should be with caution to prevent any potential phytosanitary risk from spread within or outside Canada.

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