



## Aster yellows group (16Srl), subgroups 16Srl-A and 16Srl-B, phytoplasmas associated with lettuce yellows in Texas

I.M. Lee <sup>1\*</sup>, K.D. Bottner-Parker <sup>1</sup>, Y. Zhao <sup>1</sup>, R.E. Davis <sup>1</sup> and M.C. Black <sup>2</sup>

<sup>1</sup> Molecular Plant Pathology Laboratory, USDA, ARS, Beltsville, MD 20705, USA; <sup>2</sup> Texas A&M University Research and Extension Center, Uvalde, Tx 78802, USA

\*E-mail: [ingming.lee@ars.usda.gov](mailto:ingming.lee@ars.usda.gov)

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Lettuce (*Lactuca sativa*) is an important vegetable crop grown in the United States. Lettuce yellows (previously termed lettuce aster yellows) is a devastating disease commonly found in lettuce fields and causes significant economic loss to growers, damaging 40-100% of the crop in some seasons (Thompson, 1944). The causal pathogen was not verified until a phytoplasmal aetiology was confirmed by molecular means (Lee *et al.*, 1998). In 2013, an epidemic of lettuce yellows occurred in the Winter Garden region of Texas. The infected plants were stunted with blanching and chlorosis in young heart leaves (Fig. 1). A total of thirteen samples from two different farms, including three apparently lacking symptoms from Romaine and leaf lettuce cultivars, were collected and tested for phytoplasmas. DNA was extracted from leaf veinal tissue, and nested PCRs using universal primer pair P1/16S-SR, followed by R16F2n/R16R2n and P1A/16S-SR, were performed as described previously (Lee *et al.*, 2004) to detect the presence of phytoplasmas in the samples. The taxonomic affiliation of the putative phytoplasmas detected was determined by RFLP analysis using nested PCR products (Lee *et al.*, 1998; Zhao *et al.*, 2009). A nested PCR using primer pair rpF1/rpR1 followed by rp(I)F1/rp(I)R1A as described previously (Lee *et al.*, 2004) was used to amplify a phytoplasma DNA segment (about 1.2 kb) of the ribosomal protein (rp) operon that encompassed genes *rpIV* (*rpI22*) and *rpS3* (*rpS3*); and a nested PCR using primer pair AYsecYF1/secYR1(I) followed by AYsecYF1/AYsecYR1 (Lee *et al.*, 2006) was used to amplify a phytoplasma DNA segment (about 1.3 kb) of the partial *spc* operon that includes the complete *secY* gene. Four PCR-amplified products from each of 16S rDNA, rp, and *secY* genes were cloned and sequenced as previously described (Lee *et al.*, 2004, 2006), and deposited in GenBank (Accession Nos. KF573449-KF573456; rp, KF573441-KF573448; *secY*, KF573433-KF573440).

The results indicated all 13 lettuce samples were infected with phytoplasmas. Both lettuce cultivars were infected with 16Srl-A and 16Srl-B phytoplasmas (Fig. 2). Phylogenetic analysis with 16S rRNA (Fig. 3), rp

(Fig. 4) and *secY* gene (Fig. 5) sequences confirmed the presence of two distinct lineages (Fig. 3). This is the first report confirming that lettuce yellows in Texas is associated with two distinct phytoplasma strains. The finding implicates that multiple vectors may be involved in the disease outbreak. The information on identities of associated phytoplasmas should facilitate formulating effective strategies to combat this newly emerging lettuce yellows in Texas.

### References

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Figure 1

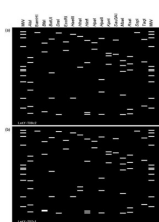


Figure 2



Figure 3

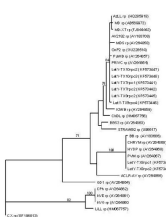


Figure 4

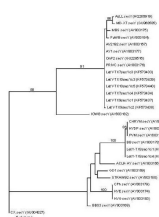


Figure 5

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