

**Characterization of the bacterial stem blight pathogen of alfalfa,
Pseudomonas syringae pv. *syringae* ALF3**

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Bacterial stem blight of alfalfa occurs sporadically in the central and western U.S. Yield losses of up to 50% of the first harvest can occur with some cultivars. Developing resistant cultivars is hampered by lack of information on the pathogen and a standard test for evaluating plant germplasm. Bacteria producing a fluorescent pigment were isolated on King's B agar from alfalfa with symptoms of bacterial stem blight from near Cheyenne, WY. The strain ALF3 was tentatively identified as *Pseudomonas syringae* pv. *syringae* based on 16S rDNA sequence and PCR amplification of *syrB* for lipodepsinonapeptide toxin production. Multilocus sequence analysis indicated that ALF3 falls within a clade containing strains of *P. syringae* pv. *syringae* with closest affinity to FF5 from pear. Comparison of a draft whole-genome sequence of ALF3 further confirmed that ALF3 most closely resembles FF5 (~96% sequence identity) and *P. syringae* pv. *aptata* DSM50252 from beet. Approximately 60 genes were unique to ALF3, including several predicted genes in the T3SS cluster such as a type III helper protein HrpZ (*Pto*) and phage-associated genes. ALF3 was highly pathogenic to snapbean pods but caused only mild symptoms on leaves of snapbean, pear, and sugarbeet. A standardized method for evaluating disease resistance in alfalfa was developed. Cultivars with fall dormancy ratings of 1 and 2 had higher percentages of resistant plants than cultivars with fall dormancy ratings of 8-11.