



First report of *Dickeya dianthicola* and *Pectobacterium wasabiae* causing aerial stem rot of potato in Michigan, USA

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Received: 21 Oct 2015. Published: 10 Feb 2016. Keywords: blackleg, *Solanum tuberosum*, tuber soft rot

Pectolytic *Dickeya* spp. and *Pectobacterium* spp. are commercially important seed-borne bacteria of potato (*Solanum tuberosum*) that cause blackleg, soft rot and aerial stem rot (Potrykus *et al.*, 2014; Stevenson *et al.*, 2001). *Dickeya* and *Pectobacterium* spp. have been recovered from potato production fields in parts of the USA (Dickey, 1979; Ma *et al.*, 2007) but a better understanding of their distribution is needed. Aerial stem rot can cause economic losses due to poor/weak stands, late emergence, and in severe cases non-harvestable fields. The disease has not been a major issue in the USA until a recent outbreak in the 2015 season. A wet June resulted in a high incidence of aerial stem rot in parts of Michigan. Two potato plants exhibiting aerial stem rot from different fields were submitted to Michigan State University Diagnostic Services for pathogen identification. Single colonies of bacteria were isolated from symptomatic tissue on nutrient-broth yeast (NBY) agar. The BIOLOG OmniLog GEN III microplate system with the associated software and database was used to identify the putative pathogenic bacteria. Results from the BIOLOG analysis identified two bacteria as *Dickeya* sp. and *Pectobacterium* sp.

DNA analysis of the bacteria was done with 16S rRNA, aconitase (*acnA*) and malate dehydrogenase (*mdh*) coding sequences as described previously (Yap *et al.*, 2004). Partial sequences of 16S rRNA (1,219 bp) and *acnA* (407 bp) genes (GenBank Accession Nos. KT626463 and KT932317, respectively) showed 99% shared identities with *D. dianthicola* strain MAFF 311044 and 302984, respectively. The *mdh* sequence (588 bp) (KT932314) shared 99% identity with sequences from *D. dadantii* strain 3937. Additionally partial sequences of 16S rRNA (1,276 bp), *acnA* (404 bp) and *mdh* (563 bp) genes (KT626462, KT932316 and KT932315, respectively) showed 99% identities with *Pectobacterium wasabiae* strain WPP163. Maximum parsimony analysis (Yap *et al.*, 2004) using concatenated 16S rRNA and *mdh* sequences from this study and *Dickeya* spp. and *Pectobacterium* spp. sequences previously deposited in GenBank clustered strain Dd2820 and Pw3230 with other *D. dianthicola* and *P. wasabiae* strains, respectively.

Five seven-week-old cv. 'Russet Norkotah' potato plants were wound-inoculated by inserting a sterile 18-gauge needle just above a central leaf axil at a depth of 1 mm. A 100 µl drop of inoculum (10⁶ cfu/ml) was placed on the wound. Plants were exposed to a 24 h leaf wetness period (90 to 100% relative humidity in a mist chamber) until symptom expression

(Figs. 1, 2), and lesions were measured. All three inoculated plants exhibited blackening of the stem and in advanced stages, drying and cracking (Figs. 1, 2). Upon desiccation, the lesions became shriveled and turned dark brown to black. Water-inoculated controls were non-symptomatic (Figs. 1, 2). The reisolated bacteria caused pitting on crystal violet pectate agar (Hélias *et al.*, 2012) and exhibited the same morphology as original cultures on NBY, and were confirmed as *D. dianthicola* using 16S rRNA and *acnA* coding sequences, and *P. wasabiae* using rRNA, *acnA* and *mdh* coding sequences, fulfilling Koch's postulates. To our knowledge, this is the first report of *D. dianthicola* and *P. wasabiae* causing aerial stem rot of potato in Michigan.

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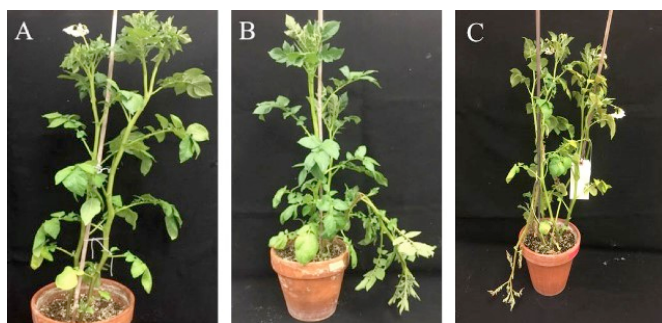


Figure 1

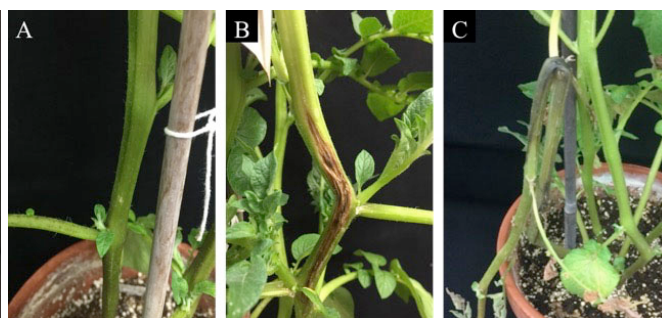


Figure 2

To cite this report: Rosenzweig N, Steere L, Kirk WW, Mambetova S, Long C, Schafer R, Dangi S, Byrne J, 2016. First report of *Dickeya dianthicola* and *Pectobacterium wasabiae* causing aerial stem rot of potato in Michigan, USA. *New Disease Reports* **33**, 10.

<http://dx.doi.org/10.5197/j.2044-0588.2016.033.010>

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