New Disease Reports

First report of pod wart disease of peanut caused by *Streptomyces* spp. in the Western Hemisphere

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In August 2018 pods with wart-like symptoms similar to those reported by Kritzman *et al*. (1996) were collected from peanut (*Arachis hypogaea* cv. Georgia-06G) plants at the University of Georgia, Bowen Farm, Tifton, Georgia, USA (Fig. 1). Putative pathogenic *Streptomyces* spp. were isolated from peanut pericarp as previously described (Wanner, 2004). Individual bacterial colonies originating from a single wart-like lesion were spread onto water agar media. Plates were incubated at 30°C for 48 hrs, and bacteria from a single colony were picked with a sterile inoculation loop and transferred to obtain pure culture.

Bacterial colonies were grey in colour with aerial and substrate mycelium similar to typical pathogenic strains of *Streptomyces* spp. (Loria *et al*., 1997). Scanning electron and light microscopy revealed structures typical of *Streptomyces* spp., that is rough, cylindrical spores (Figs. 2-3) in flexuous chains. Genomic DNA was extracted from cultures grown on oat bran agar media at 30°C. Mycelia were scraped from the agar surface with a sterile toothpick and DNA extracted using the DNeasy Power Soil Kit (Qiagen Inc., USA) according to the manufacturer's instructions. DNA-based analyses targeting the 16S rRNA, recombinase A (*recA*), RNA polymerase b-subunit (*rpoB*) and thaxtomin synthase (*txtAB*) were used. Partial sequences of the 16S rRNA (1,219 bp), *recA* (913 bp), *rpoB* (994 bp) and *txtAB* (385) genes were amplified using primers previously reported (Wanner, 2006; Guo, 2008). PCR products were sequenced and submitted to GenBank (Accession Nos. MK630207.1, MK737925 and MK749839 respectively). BLASTn analysis of 16S rRNA (LC207997.1), *recA* (MF925465.1) and *rpoB* (KX503547.1) sequences revealed 99% to 100% identity with *Streptomyces* spp. and had the pathogenicity gene encoding *txtAB*.

Three plants of cv. Talbert Small Red were each inoculated with a 50 ml (10^6 CFU/ml) bacterial suspension using a soil drench method to confirm pathogenicity. Non-inoculated plants served as controls. Plants were grown in the greenhouse under a 16 hour photoperiod. Peanuts were harvested 133 days after planting and assessed for the presence of wart symptoms (Fig. 4). Pods recovered from non-inoculated controls were asymptomatic. *Streptomyces* spp. were re-isolated and recovered from inoculated symptomatic peanut pericarps as described above. The recovered bacteria were subsequently identified using the 16S rRNA, *recA* and *rpoB* coding sequences. Thus, the host plant inoculation coupled with DNA sequencing analyses of re-isolated bacteria fulfilled Koch's postulates.

Peanut wart has been reported in Israel (Kritzman *et al*., 1996) and South Africa (de Klerk, *et al*., 1997). However, to our knowledge, this is the first report of *Streptomyces* spp.causing pod wart disease of peanut in the United States and the Western Hemisphere.

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Figure 1

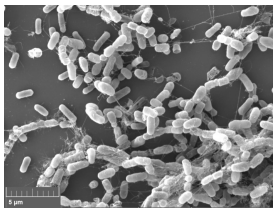

Figure 2

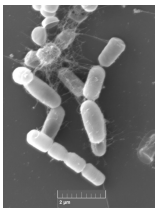

Figure 3

Figure 4

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