New Disease Reports

First report of *Pineapple mealybug wilt associated virus-1* in Ecuador

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In Ecuador, where pineapple represents one of the most important export commodities, virus testing has been neglected. In July 2014, a total of twenty MD2 hybrid pineapple plants showing virus-like symptoms (Fig. 1) were collected from a commercial planting located at the border of Santo Domingo and Los Rios provinces, where most of the pineapple in Ecuador is grown. Double-stranded RNA (dsRNA) was extracted from 20 g batches of leaves as described by Morris & Dodds (1979). The dsRNA was heatdenatured and used as template to generate cDNA libraries using anchoredrandom primers. Sequencing of randomly amplified PCR products revealed the presence of Pineapple mealybug wilt associated virus 1 (PMWaV-1). Partial sequences for the helicase (Hel) and the coat protein (CP) genes were re-sequenced using specific primers and deposited in GenBank under Accession Nos. KP728914 and KP728915, respectively. Diagnostic primers (PMWaV-1_EcF: 5'-CCAAGCGAGAGGAAGGAATGTAAG-3'; PMWaV-1_EcR: 5'-CTCTGAATAATTGCCGAAACTCGG-3') were designed to amplify a 217 nt fragment of the Hel gene. Upon confirmation of specificity, the primers were used to test 150 plants (75 symptomatic and 75 asymptomatic) collected from Santo Domingo and Los Rios provinces, where the MD2 hybrid is grown, and Guayas province, where 'Smooth Cayenne' is the cultivar of choice. Mealybugs (Dysmicoccus spp.) were prevalent in sampled fields. PMWaV-1 was detected in 33% and 77% of asymptomatic and symptomatic MD2 plants, respectively. In the cultivar

'Smooth Cayenne', the virus was detected in 37% and 65% of asymptomatic and symptomatic plants, respectively. These findings suggest the presence of additional virus(es) possibly associated with the observed symptoms, as has been reported elsewhere (Hernandez-Rodriguez *et al.*, 2014). Sequence alignments showed that PMWaV-1 from Ecuador is related most closely to Hainan isolate from China (KJ872494.1) with nucleotide identities of 99% and 98% for the CP and Hel genes, respectively. To the best of our knowledge, this is the first report of PMWaV-1 infecting pineapple in Ecuador.

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Figure 1

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