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Stylet penetration activities of *Diaphorina citri* associated with transmission of *Candidatus* Liberibacter asiaticus

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The electrical penetration graph (EPG) technique was used to determine *Diaphorina citri* stylet penetration activities associated with *Candidatus* Liberibacter asiaticus (Las) acquisition and inoculation in citrus. In a first experiment, healthy *D. citri* adults were connected to the EPG system and placed on Las-infected plants. Probes were artificially terminated after the following stylet penetration periods and waveforms: I) 20 min in waveform C (pathway phase through epidermis and parenchyma); II) C + 30 s in D (first contact with phloem tissue); III) C+D + 70 s in E1 (penetration and possibly salivation/ingestion in the phloem sieve elements); and IV) C+D+E1+ 1 h in E2 (phloem sap ingestion). The insects were tested for Las infectivity by real-time PCR 3 wks later. In a second experiment, 3rd-instar nymphs were first submitted to an acquisition access period of 2 wks on Las-infected plants and then connected to the EPG system on healthy citrus seedlings during the same stylet penetration periods of the first experiment. *D. citri* acquired the pathogen only after penetration in the phloem sieve elements and mostly during waveform E2 (27 infective insects of 54 tested). Only 2 of 52 insects exposed to infected plants until waveform E1 (treatment III) were positive. In the second experiment, transmission (by 9 of 50 insects tested) was observed only by psyllids allowed to perform E2 on healthy seedlings. Overall, the data show that both acquisition and inoculation occur during the phloem phase, primarily during sustained sap ingestion (E2). This information is important for designing efficient control tactics aimed at preventing Las transmission by *D. citri.*
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