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Aphid transmission and alternate hosts of Passiflora chlorotic mottle virus in Uganda

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Abstract

This research established aphid vectors and alternate hosts of Passiflora chlorotic mottle virus in Uganda. Aphids and host plants were collected from farmers' fields in major passion fruit growing districts in Uganda. Transmission tests were carried out in passion fruit and *Nicotiana benthamiana* using the aphids. Double antibody sandwich enzyme-linked immunosorbent assays (DAS-ELISA) and reverse transcription polymerase chain reactions were carried out to confirm virus presence. Results reveal cowpea aphid (*Aphis craccivora*), bean aphid (*Aphis fabae*) and green peach aphid (*Myzus persicae*) as efficient vectors of the virus. Common bean, cowpea, black jack, spiny amaranth and spinach were also good alternate hosts for the virus.

Key words: Epidemiology, ELISA, passion fruit woodiness disease, RT-PCR, viral disease

Résumé

Cette recherche a établi les vecteurs de pucerons et en alternance avec des porteurs des virus tachetés de Passiflora chlorotique en Ouganda. Les pucerons et de plantes porteuses ont été recueillis dans les champs des agriculteurs dans les principaux districts de production des fruits de la passion en Ouganda. Les tests de transmission ont été effectués sur les fruits de la passion et *Nicotiana benthamiana* en utilisant les pucerons. Des tests d'insertion de double anticorps de dosage immuno-absorbant -enzymatiques (DAS-ELISA) et la transcription inverse des réactions en chaîne de la polymérase ont été effectués pour confirmer la présence du virus. Les résultats révèlent les pucerons de niébé (*Aphis craccivora*), puceron du haricot (*Aphis fabae*) et le puceron de pèche vert (*Myzus persicae*) comme des vecteurs efficaces du virus. Le haricot, le niébé, l'amarante épineuse et les épinards sont également de bons porteurs alternatifs pour le virus.

Mots clés: épidémiologie, ELISA, maladie de fruit de la passion, RT-PCR, maladie virale

Background

In Uganda, viral diseases causes between 40-100% yield loss and total crop loss in some cases (Wasilwa *et al*., 2004). The diseases have been associated with a putative novel species the Passiflora chlorotic mottle virus (PaCMV) (Ochwo-Ssemakula *et al*., 2012).

This virus causes a diversity of symptoms including: chlorosis, mosaic, fruit woodiness and vein clearing on different passion fruit types (Ochwo-Ssemakula, 2008). It belongs to the family *Potyviridae*, genus *Potyvirus* where viruses that cause similar symptoms occur, including: *Passion fruit woodiness virus*, *Cowpea aphid borne mosaic virus* and *East Asian Passiflora virus* (Ochwo-Ssemakula, 2008). Among other mechanisms, these viruses are transmitted by aphids in a non-persistent manner (Shukla *et al.,* 1998). Genomic information for PaCMV indicates the species as being aphid transmissible (Ochwo-Ssemakula, 2008). However, being a novel species, there was limited information on potential aphid vectors of the virus and alternate hosts in Uganda. This study was under taken to determine the aphid species and alternate host plants associated with PaCMV in passion fruit fields in Uganda. Since passion fruit is not a preferred host (Novaes & Rezende, 2005), this information will contribute towards a better understanding of the epidemiology of viral disease on passion fruit and the development of appropriate management strategies.

Literature summary

Passiflora chlorotic mottle virus is similar to and yet distinct from *Passion fruit woodiness virus* (PWV), *Cowpea aphid borne mosaic virus* (CABMV) and other viruses causing passion fruit woodiness disease worldwide such as *Cucumber mosaic virus* (CMV) and *East Asian passiflora virus* (EAPV) (Ochwo-Ssemakula, 2008). Viruses belonging to the genus Potyvirus are naturally transmitted by several species of aphids such as *Myzus persicae*, *Aphis gossypii*, *A. spiraecola* and *Toxoptera citricidus* in a non-persistent manner, and through grafting and mechanical inoculation (Fischer and Rezende, 2008). Aphid-borne non-persistent viruses (ABNPVs) are better managed by targeting the source of inoculum (Irwin, 2009) that includes the planting material, volunteer crops, and alternative hosts of both the vector and viruses. Effective disease management practices include: selection of resistant plants, use of pesticides, pre-immunization with mild strains of the virus and the adoption of cultural practices that can minimize the incidence and dissemination of viral diseases (Trevisan *et al*., 2006).

Management of passion fruit viral diseases should target the source of the viruses and vectors, in this case the alternative hosts, both within and outside the field. Such management options may present a challenge considering that passion fruit viruses and aphid vectors have a diverse host range (Fischer & Rezende, 2008). Strategies developed for similar viral diseases have, however, proved successful.

Study description

Surveys were conducted in Mukono and Mbale, which are major passion fruit growing districts in Uganda. These districts have also been found to harbor isolates of Passiflora chlorotic mottle virus with different genetic profiles (Ochwo-Ssemakula, 2008). Ten farmers were selected in each district. On each farm, five water pan traps were placed along the length of the field to trap aphids. Aphid samples were also collected from five growing points of each vine and host plants in the field and within a radius of 100m. Samples were either preserved in 70% alcohol for identification at the Makerere University Agricultural

Research institute Kabanyolo (MUARIK) or cultured on host plants in a screen house. Species identification was done under a digital Dino-lite microscope with the help of existing keys based on morphological features according to Blackman and Eastop (2000). Distinguishing features included: body color, antennal tubercles development and placement, cornicle length relative to cauda and caudal hairs. Alternate hosts were identified using a plant identification guide (Castner, 2005).

Transmission tests were carried out for all the identified aphid species to passion fruit and *N. benthamiana* as described by Wang et al. (1996). Data were collected for disease incidence and severity according to Hahn et al (1980). Leaf samples were collected and tested for presence of PaCMV using DAS-ELISA) with antisera for detecting Potyviruses and PaCMV, and confirmed using RT-PCR with Potyviridae primers (Chen *et al*., 2003).

Research application

Four species of aphid were sampled from the study sites: *Aphis craccivora, Aphis fabae*, sowthistle aphid (*Hyperomyzus lactucae*) and *Myzus persicae* (Table 1). ELISA tests detected presence of PaCMV in leaf samples as shown in Table 2. *Aphis craccivora, Aphis fabae, Hyperomyzus lactucae* transmitted the virus to varying degrees (Table 2).

Table 1. Aphid species sampled from passion fruit farmers' fields in Mukono and Mbale districts in Uganda.

Aphid species	Aphids sampled			
	Mukono		Mbale	
	No.	Percentage (%)	No.	Percentage (%)
Aphis craccivora	13	4.38	206	45.47
Aphis fabae	21	7.07	15	3.31
Myzus persicae	150	50.51	0	0
Hyperomyzus lactucae	113	38.04	232	51.21
Total	297	100	453	100

Table 2. Detection of PaCMV in three aphid species from passion fruit farmers' fields in Uganda.

Aphid species	Symptomatic plants (%)	Mean ELISA readings (nm)
Aphis craccivora	53.8	0.2457
Aphis fabae	23.3	0.2193
Hyperomyzus lactucae	22.9	0.169
*control	0	0.0515
Total	100	

Preliminary results from RT-PCR confirm presence of PaCMV in passion fruit, *N. benthamiana* and aphid samples.

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