



## TRABALHOS APROVADOS PARA APRESENTAÇÃO EM FORMA DE PÔSTER

### 180 - ÁREA: VEGETAL E INVERTEBRADOS

#### FIRST REPORT OF PASSION FRUIT GREEN SPOT VIRUS IN THE STATE OF MATO GROSSO, BRAZIL

CAMILA CHABI DE JESUS, PEDRO LUIS RAMOS GONZÁLEZ, PATRICIA DE FARIA TOLEDO, RICARDO HARAKAVA, ALINE TASSI, RAFAEL BURILLE, SUZINEI SILVA OLIVEIRA, HERMES PEIXOTO SANTOS FILHO, ELLIOT WATANABE KITAJIMA, JULIANA DE FREITAS-ASTÚA

<sup>1</sup> ESALQ - Escola Superior de Agricultura Luiz de Queiroz/USP, <sup>2</sup> IB - Instituto Biológico, <sup>4</sup> CUSC - Centro Universitário São Camilo, <sup>5</sup> Yasaí - Yasaí Alimentos, <sup>6</sup> EMBRAPA Agrossilvipastoril - Empresa Brasileira de Pesquisa Agropecuária, <sup>7</sup> EMBRAPA Mandioca e Fruticultura - Empresa Brasileira de Pesquisa Agropecuária  
*millachabi@yahoo.com.br*

#### Resumo

Passion fruit green spot disease, caused by Passion fruit green spot virus (PfGSV), affects plants of genus *Passiflora* and is characterized by chlorotic or necrotic lesions on fruits, leaves and branches. In severe cases, a general dieback is observed, leading to the death of the infected plants. The disease has been only described in few areas of SP, MA, BA and DF. PfGSV is a tentative cilevirus [ss(+)RNA] transmitted by false-spider mites of the genus *Brevipalpus*. In 2016, passion fruit producers from Tabaporã and Sinop, MT, reported severe necrotic symptoms and the death of passion fruit plants. To identify the causal agent of the disease, samples were submitted to transmission electron microscopy (TEM) analyses and RT-PCR for the detection of PfGSV. *Brevipalpus* mites occurring in the symptomatic samples were also collected for their morphological characterization. Total RNA was extracted from the lesions using Trizol® and three specific primer pairs were used to detect the putative presence of both RNA1 and RNA2 molecules from PfGSV. All samples tested positive for PfGSV, yielding the expected bands in 1% agarose gels. Amplicons were sequenced and exhibited more than 98% nucleotide identity with the partial sequence of PfGSV available in the GenBank (HM002746 and HM002747). Bacilliform particles (50-70 nm x 100-120 nm) and dense viroplasm, similar of those caused by cileviruses, were observed in TEM in the cytoplasm of infected cells. No other viral particles were detected by TEM. All mites collected in symptomatic plants (10 specimens) were identified as *B. yothersi*. This study is the first report of the disease in the state of Mato Grosso, Brazil. After the identification of the disease and notification of the results, passion fruit growers were able to proceed with the correct management of the disease through the control of the mite vector in the Tabaporã and Sinop areas.

**Palavras-chave:** *Cilevirus*, *Brevipalpus* mites, RT-PCR



## TRABALHOS APROVADOS PARA APRESENTAÇÃO EM FORMA DE PÔSTER

### 402 - ÁREA: VEGETAL E INVERTEBRADOS

#### MOLECULAR DIVERSITY AND GEOGRAPHIC DISTRIBUTION OF CITRUS LEPROSIS VIRUS C IN SOUTHERN SOUTH AMERICA

CAMILA CHABI DE JESUS, PEDRO LUIS RAMOS GONZÁLEZ, MARIANE DA COSTA RODRIGUES, MATHEUS POSTSCLAM BARRO, RICARDO HARAKAVA, ELLIOT WATANABE KITAJIMA, RENATO BEOZZO BASSANEZI, JULIANA DE FREITAS-ASTÚA

<sup>1</sup> ESALQ - Escola Superior de Agricultura Luiz de Queiroz/USP, <sup>2</sup> IB - Instituto Biológico, <sup>3</sup> UAM - Universidade Anhembi Morumbi, <sup>4</sup> UNIP - Universidade Paulista, <sup>5</sup> Fundecitrus - Fundo de Defesa da Citricultura, <sup>6</sup> CNPMF - Embrapa Mandioca e Fruticultura  
*millachabi@yahoo.com.br*

#### Resumo

Cytoplasmic type of citrus leprosis disease (CL-C) is the main viral disease affecting Brazilian citrus orchards. The disease causes localized chlorotic and/or necrotic lesions and induces premature leaf and fruit drop. Citrus leprosis virus C [CiLV-C, genus Cilevirus, ss(+)RNA], is the prevalent virus producing CL-C from Mexico to Argentina and from which two viral clades are known: CRD and SJP. Extant members of the clade SJP were firstly discovered in 2015 and bioinformatic tools indicated that viral strains from the two clades were involved in putative natural recombination processes. To shed some light about the molecular epidemiology of CL-C, viruses of the two clades were surveyed in citrus orchards where the disease is widespread. Samples kept in -80°C freezer were also included in the study. A total of 84 samples [Brazil (66), Paraguay (3), Bolivia (1) and Argentina (4)] of sweet orange plants showing typical symptoms of CL and collected during the period 2003-2017 were tested by RT-PCR for the confirmation of CiLV-C presence and the specific identification of the occurring CiLV-C strains. Generated amplicons were independently sequenced by the Sanger method. Results indicated the unequivocal presence of CiLV-C in all samples, confirming that it is the prevalent virus associated to CL in Southern South America. Strains of the clade SJP were detected in samples from Argentina (1), Paraguay (1) and Brazil (45) collected since 2010. Forty-three of the Brazilian samples were from orchards of the state of SP and two from MG. Moreover, CRD strains were detected in SP (26), MG (2), PR (1), ES (1), RS (1), DF (1), and in Argentina (4), Paraguay (4) and Bolivia (1). Remarkably, viruses from the two clades were simultaneously detected in 19 samples from SP, indicating the occurrence of mixed infections in isolated fruit lesions. These results give a natural contextual support for the recombinant processes previously predicted, alert for the epidemiology consequences resultant from the interaction of viral mixed infection with their mite vectors and, additionally, offer insights into the evolutionary forces driving the distribution and diversity of CiLV-C population along the region harboring the largest extension of sweet orange orchards of the world. Financial Support: FAPESP 2016/01960-6 and 2014/08458-9.

**Palavras-chave:** *Cilevirus*, CiLV-C, *Brevipalpus* mites



## TRABALHOS APROVADOS PARA APRESENTAÇÃO EM FORMA DE PÔSTER

### 406 - ÁREA: VEGETAL E INVERTEBRADOS

#### TRANSCRIPTOME OF THE CITRUS LEPROSIS VIRUS C - BREVIPALPUS YOTHERSI INTERACTION

THAIS ELISE SINICO, GABRIELLA DIAS ARENA, ELLIOT WATANABE KITAJIMA, MARCOS ANTÔNIO MACHADO, VALDENICE MOREIRA NOVELLI, JULIANA FREITAS-ASTÚA

<sup>1</sup> ESALQ-USP - Escola Superior de Agricultura "Luiz de Queiroz", Universidade de São Paulo, <sup>2</sup> UNICAMP - Universidade Estadual de Campinas, <sup>3</sup> CCSM-IAC - Centro APTA Citros "Sylvio Moreira" – IAC, <sup>4</sup> EMBRAPA - Embrapa Mandioca e Fruticultura, <sup>5</sup> Instituto Biológico - Laboratório de Bioquímica Fitopatológica, Instituto Biológico

*thais.sinico@usp.br*

#### Resumo

*B. yothersi* tenuipalpid mites are the main vectors of *Citrus leprosis virus C* (CiLV-C), the causal agent of citrus leprosis disease, currently the main viral disease of citrus in Brazil. The disease is characterized by symptoms that include local chlorotic and necrotic lesions restricted to the mite feeding sites, leading to leaf and fruit drop, and demanding high costs with control measures. However, there are very few studies on the biology of *B. yothersi* mites and their interaction with CiLV-C. Thus, through the study of the mite vector transcriptome, we investigated the differential gene expression (DGE) profiles between aviruliferous and viruliferous populations using the RNA-Seq technique. For this, mite populations were reared onto sweet orange fruits (healthy and symptomatic for CiLV-C), and a pool of 500 mites were collected with four replicates/treatment for the extraction of total RNA. Then, molecular analyses were performed by RT-PCR to certify the status of each treatment, and the samples were sequenced by a HiSeq 2500 Sequencing System – Illumina. The data were analyzed with R language and Bioconductor software packages. Reads were mapped in the *B. yothersi* genome and analyses of DGE were performed. We identified 841 differentially expressed genes in *B. yothersi* viruliferous population, with 711 transcripts being up-regulated and 130 down-regulated. Functional analyses were performed using Blast2GO and the NCBI database, targeting similar proteins in *Tetranychus urticae* mites and others arthropods. Detoxification-related genes, such as acetylcholinesterase and cytochrome P450 monooxygenase were found, as well as possible genes involved in virus-vector interaction. The most expressed genes and those putatively important for the leprosis pathosystem were selected and are being validated by qPCR, aiming to elaborate a model of the virus-mite vector interaction. Financial Support: CAPES, FAPESP (Process number 2014/08458-9)

**Palavras-chave:** Brevipalpus mites, Cilevirus, virus-vector interaction, RNAseq