

Sittichai Boonrawd 2010: Isolation, Identification and Partial Nucleotide Sequence of a Flexuous Rod Virus Causing Yellow Spot Mosaic of Purple Passionfruit. Master of Science (Genetic Engineering), Major Field: Genetic Engineering, Interdisciplinary Graduate Program. Thesis Advisor: Assistant Professor Pissawan Chiemsombat, Dr.Agr. 73 pages.

Purple passionfruit (*Passiflora edulis*) No.2 which is widely grown in the Royal Project extension area in Chiangmai Province is severely affected by woodiness disease caused by a virus. The disease causes low yield and decreases the quality of the fruits. It is necessary to identify the causal virus and inspect mother plants in order to produce virus free stocks. Twenty diseased passionfruit plants investigated in this study showed two types of symptoms e.g. severe leaf blistering with yellow spot mosaic, and dark and light green mosaic leaf with distortion. The virus was isolated from infected leaves by mechanical inoculation on the assay plant, *Chenopodium quinoa* and *C. amaranticolor*. Chlorotic local lesions developed on inoculated *Chenopodium* leaves were singly transferred to passionfruit seedlings for virus propagation. The passionfruit plant developing yellow spot symptom was selected for further study and called SY2 isolate. Morphological and ultrastructural study, and partial nucleotide sequencing were performed for virus identification. Dip preparation of SY2 leaves revealed flexuous rod shaped particles with modal length of about 11.3 x 642 nm. Pinwheel, tubular and scroll inclusion bodies were readily detected in cytoplasm of the infected leaf cells. Host range of SY2 isolate included cowpea; *Vigna sinensis*, yard long bean; *Vigna sesquipedalis*, white bean; *Bruguiera cylindrica* and kidney bean; *Phaseolus vulgaris* cv. Yamashiro. A fragment of viral 3' proximal genomic RNA was amplified by using RT-PCR, cloned and sequenced. This fragment is 1,563 nucleotide (nt) long, comprising 630 nt of partial Nib gene, 819 nt of entire coat protein (CP) gene, and 111 nt of partial 3' untranslated (UTR) region. ClustalW analysis of amino acid sequence revealed that SY2 isolate belonged to a potyvirus member with 87% highest identity to *Telosma mosaic potyvirus* (TeMV), and contained a chain of 17 unique amino acid sequence at positions 18-35 of CP-N terminal. In addition, SY2 partial 3' UTR sequence was 81% identity to TeMV. Phylogenetic tree reconstructed by CP amino acid sequence showed pairing of SY2 isolate with TeMV in Vietnam, and formed a cluster with potyvirus species causing woodiness disease of passionfruit in Australia, ornamental potyviruses in America, and bean potyviruses in China and South Africa. The second cluster contained distinct passionfruit woodiness viruses from Taiwan, Japan, soybean mosaic virus, and several legumes potyviruses. From these results, SY2 potyvirus causing woodiness disease of purple passionfruit No.2 is considered to be a closed strain to TeMV and closed species to PWV-Aus. The name passionfruit woodiness virus yellow spot strain; PWV-YS is proposed.

---

Student's signature

---

Thesis Advisor's signature

