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Isolation of A Tetravirus from *Setothosea asigna* Larvae Infected with An Epidemic Disease In Oil Palm Plantation In South Sumatra, Indonesia

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Introduction

Larvae of nettle caterpillar, *Setothosea asigna, Setora nitens, Darna trima and Darna Bradleyi* (Lepidoptera ; Limacodidae), are sympatric and coseasonal; limacodid moths in Southeast Asia and being important defoliator of oil palm and coconuts palms. In severe infestations, *Setothosea asigna* (*S.asigna*) larvae consume all foliage and leave only mid-rib of the frond, causing setback of fruit production for a year. Thus, *S. asigna* become an economically important defoliator in oil palm (*Elaeis guineensis*) plantation in north-eastern Sumatra, Borneo and western Malaysia. Recurring outbreak of *S. asigna*, singly, or together with heterospecific limacodids, provoked application of broadspectrum insecticides. However, recently, the environmental pollution by agricultural chemical becomes a big social problem in many countries, and Indonesia is no exception.

Recently, natural epidemics among *S. asigna* larvae suspected of viral infection have been occurred in some oil palm fields in Indonesia. These infectious diseases resulted in decrease of pest population below an economic threshold in those epidemic areas. Suggesting the possibility of an effective control of *S.asigna* by viral pesticides isolated from epidemic field. However, little is known yet about the causing agent of natural epidemic disease among *S. asigna* observed in Indonesia

Result and Discussion

Diseased larvae of *S. asigna* were collected in an epidemic field and used as a starting material for purifying viruses. The existence of RNA with the sequence homology at least partly to the *RdRp* sequence of TaV was demonstrated by RT-PCR followed by sequence analysis. This result suggested that TaV or TaV-related virus is existed in the infected larvae. Purification of virus particles was then performed by a method described for Dendrolimus punctatus tetravirus (DpTV) purification by Fuming Yi *et al,* (2005). On a sucrose density gradient of partially purified viral fraction, only one white band was observed. The white band was collected, precipitated by ultra centrifugation, and examined with electron microscopy.

Negatively stain electron microscopy showed that the fraction contained numerous numbers of non-enveloped, spherical virus-like particles with about 40 nm in diameter. The morphological characteristics of the purified particles resembled those of members of the family *Tetraviridae*. The cDNAs against RNAs extracted from the purified viral particles were generated with random primers and sequenced using an automatic sequencer. The viral RNA showed a similarity of about 99% in total with that of *Thosea asigna* virus (TaV), a member of the genus *Betatetravirus* previously reported.

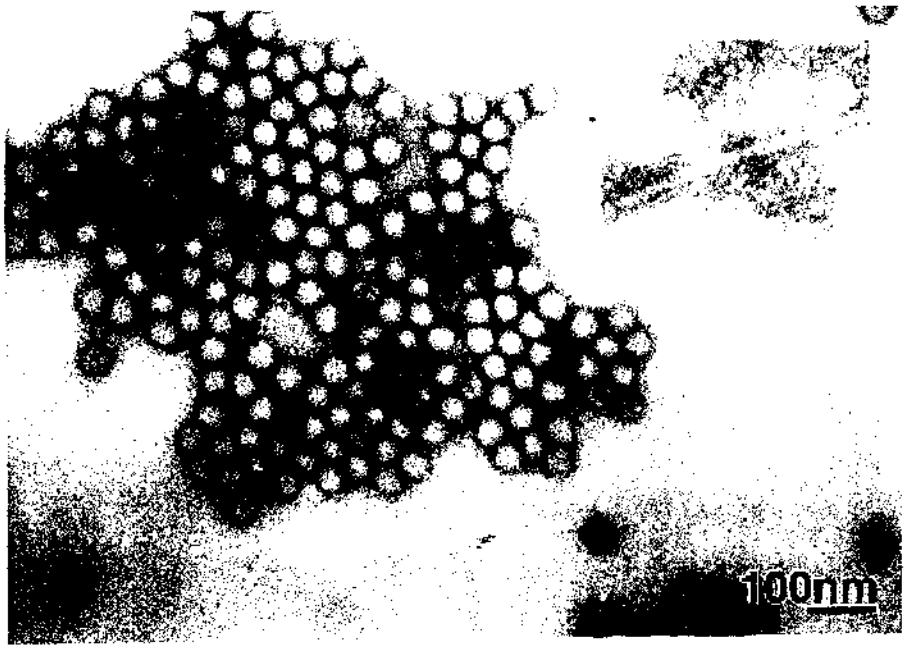

Figure 1. Negative stain electron microscopy of viral particles purified by sucrose density gradient centrifugation. Rod shaped particles (t) are tobacco mosaic viruses (TMVs) used as an indicator

Conclusions

In conclusion, *Thosea asigna* Virus was isolated from *S. asigna* larvae infected with an epidemic disease in an oil palm plantation in South Sumatra. This result suggesting the possibility that TaV isolated in this study would be a useful resource for a biological control of *S. asigna* in oil palm plantations.

References

Entwistle, P.F. 1987. Virus diseases of limacodidae. In slug and nettle caterpillar : The Biology, taxonomy and control of the limacodidae of economic importance on palm in south-east asia (pp. 213-221). Holloway. Walling ford : C.A.B. International.

Hendry, D.A., & D.K. Agrawal. 1994. Tetravirus. Encyclopedia of Virology.

Hendry, D.A., Johnson, J.E., Rueckert, R.R., Scotti, P.D & Hanzlik, T.N. 1995. Family Tetraviridae. In Virus Taxonomy: six Report of the International Committee on Taxonomy of Viruses (pp. 372-375). New York : Springer-verlag.

Pringle, M.F., Gordon, K.H.J., Hanzlik, T.N., Kalmakoff, J., Scotii, P.D. & Ward, V.K. 1999. A novel capsid expression strategy for Thosea asigna virus (Tetraviridae). Journal of general Virology, 80, 1855-1863. Retrieved from http://vir.sgmjournals.org/

Reinganum, C., Robertson, J.S., & Tinsley, T.W. 1978. A new group of RNA virus from insects. Journal of general Virology,. 40, 195-202. Retrieved from http://vir.sgmjournals.org/

Yi, F. M., Zhang, J. M., Yu, H. Y., Liu, C. F., Wang, J. P. and Hu, Y. Y. 2005. Isolation and identification of a new tetravirus from *Dendrolimus punctatus* larvae collected from Yunnan Province, China. Journal of generalVirology,86: 789-796. Retrieved from http://vir.sgmjournals.org/