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Using electron microscopy to uncover latent tobamovirus

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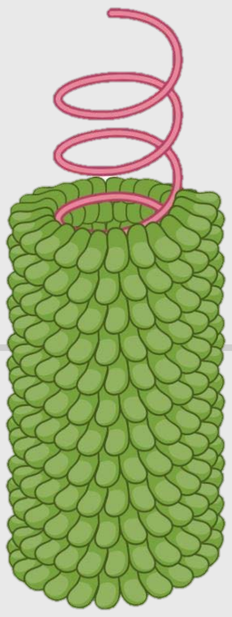
Tobamoviruses

- Family: *Virgaviridae* (contains 37 species)
- Properties: +ssRNA viruses, rod shaped
- Transmission: mechanical



Latency

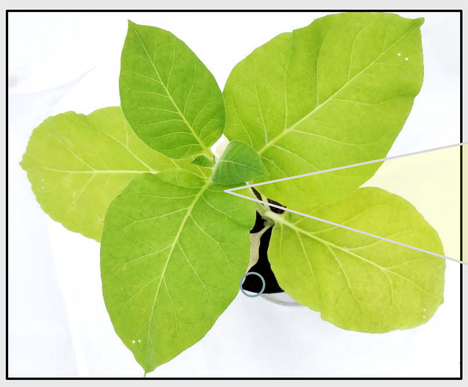
The virus can replicate and move systemically but does not cause disease[1].

Objective

- Detection and characterization of asymptomatic plant virus isolated from Hoya spp.
- Hoya tobamovirus-2 (HoTV-2) is a new tobamovirus of ornamental plant reported in Germany[2].
- Asymptomatic in original host plant

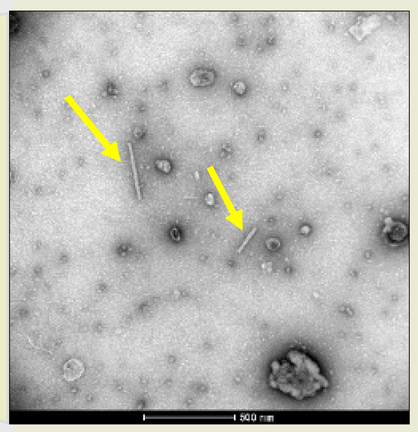
Results

1. HoTV-2 was asymptomatic on experimental host plants

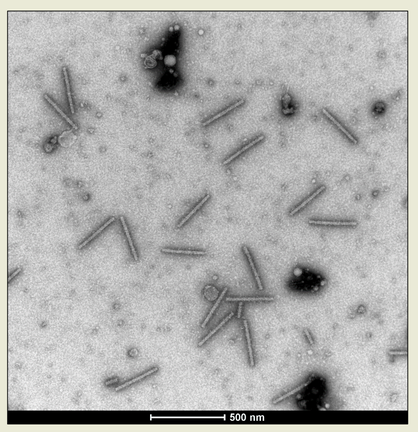


N. tabacum cv. Samsun (nn) infected with HoTV-2

2. Rod shaped virions were observed in the asymptomatic plants. Youcai mosaic virus (YoMV) reacted and trapped 20X more virions on EM grid

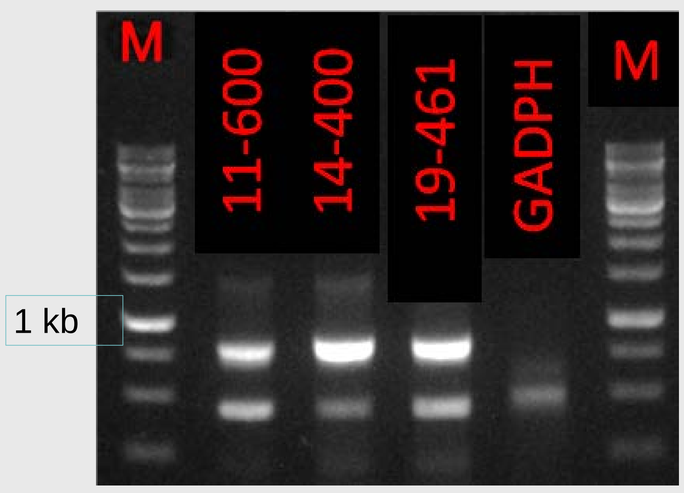


Overview of HoTV-2 virions observed under EM from the asymptomatic N. benthamiana leaf.



Immunoassay using a close relative antisera trapped virions effectively on grid. antisera.

3. Tobamo-generic primers were used to confirm presence of a tobamovirus in asymptomatic plants[3].



4. Sanger sequencing of RT-PCR products showed presence of a new species as per species demarcation criteria for tobamoviruses

AKR0022209	TEM11-600	Tobuni 1-f	72% Hoya chlorotic spot virus sequence	KX434725.1
AKR0022210	TEM14-400	Tobuni 1-f	65% Hoya chlorotic spot virus sequence	KX434725.1
AKR0022211	TEM19-461-466	Tobuni 1-f	68% Hoya chlorotic spot virus sequence	KX434725.1

Conclusion

- YoMV antisera (DSMZ AS 1496) can be used for detection and trapping of HoTV-2
- Generic tobamo primers can be used for detection of HoTV-2

Outlook

Future studies will be focused on:
- Quantitative analysis of HoTV-2 in single/multiple infections
- Ultrastructure of HoTV-2 in single/multiple infections

Acknowledgement

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References

1.Ilyas et al. To Be Seen or Not to Be Seen: Latent Infection by Tobamoviruses. *Plants* **2022**, *11*, 2166. https://doi.org/10.3390/plants11162166
2.Gaafar et al. A new tobamovirus infecting *Hoya* spp. *New Dis. Rep.* **2020**, *42*, 10, doi:10.5197/j.2044-0588.2020.042.010.
3.Letschert et al. Detection and differentiation of serologically cross-reacting tobamoviruses of economical importance by RT-PCR and RT-PCR-RFLP. *J. Virol. Methods* **2002**, *106*, 1–10.

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