Molecular Variability within the coat protein gene of Hop Mosaic and Hop Latent carlaviruses

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Australian production is free from major fungal diseases and pests that cause significant losses in hop-growing areas around the world. The main pathological cause of yield loss is infection by viruses. Three viruses are commonly found in Australian hop gardens, *Hop latent carlavirus* (HpLV), *Hop mosaic carlavirus* (HpMV) and *Apple mosaic ilarvirus* (ApMV).

Two distinct strains of ApMV infecting hop have been identified through serological relationships and by sequence analysis of the coat protein (CP) gene. Whilst possible symptom variants have been observed, there is no serological or molecular data to support variation within the common hop carlaviruses. Here we present partial coat protein sequence data for Australian isolates of HpMV and HpLV and show evidence for two distinct strains of HpMV.

Methods

Sequences of single isolates of HpLV (accession no. E13365) and HpMV (accession no. AB051109) sourced from Japanese hop have been published. Primers derived from these sequences were used for PCR amplification of part of the CP gene of both HpLV (6 isolates) and HpMV (4 isolates) from infected hops in commercial gardens at Forrester River, Gunns Plains and Bushy Park. Amplicons were directly sequenced in both directions to confirm sequence identity using internal primers (Figure 1). Sequences (nucleic acid; na and predicted amino acid; aa) were aligned and compared to the published sequence. Hydrophobicity profiles were generated for the consensus amino acid sequences.



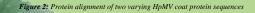
Figure 1: Schematic for HpLV and HpMV PCR amplification and sequencing

Hop latent virus

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The partial CP sequences from the Australian HpLV isolates shared near identity with the published Japanese isolate (96-97% na homology). The few base pair substitutions observed were generally conserved across the Australian isolates, however, these substitutions did not alter the predicted aa sequence of the CP.

AA residue #	\$ 3	8	11		22 24	86	95 98
HpMV CP1	LIQIM	S G N T 1	TGKI		PKDA	ETKVQITNSSLE	GRPALALTTDMRK
HpMV CP2	ĸ	S	λ		Q Y G	G	т А
AA residue #	255			270	275	285	303
HpMV CP1	A V Q P L	DGIVR	R P T P F	EKLAHS	THKDI	ALRKAN RNOHFGNMD	VEVTGGRSGPEIVRDYSK



Hop mosaic virus

The four Australian HpMV isolates formed two distinct groupings based on na and predicted aa sequence. Two isolates had a high level of na homology to the Japanese isolate (97.3-98.0%), with the other two showing distinct differences (85.0-85.4% na homology to the Japanese isolate). The two divergent isolates were sourced from a cultivar reference collection at Bushy Park that included genotypes introduced from around the world that may have been infected with HpMV when imported. The two isolates with near identity to the Japanese isolate were sourced from distinct commercial gardens in Forrester River. When a predicted aa alignment was performed (311 aa residues), differences were noted at the start (first 100) and end (last 60 residues) of the partial CP sequences (Figure 2). This is reflected in the hydrophobicity profiles of the two viruses (Figures 3 & 4).



Figure 3: Hydrophobicity profiles of two varying HpMV coat protein sequences

HpMV CP 1

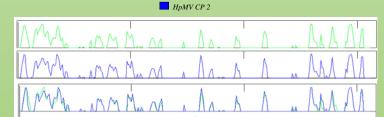


Figure 4: Hydrophobicity profiles of two varying HpMV coat protein sequences





Conclusions

Sequence data from the coat protein genes of a collection of Australian isolates of HpLV and HpMV has been compared to published sequences from Japan. All HpLV isolates were highly homologous to each other and the Japanese isolate. In contrast, two distinct groups of HpMV were suggested with both groups present in Australian hop. Further work is required to ascertain if these sequence variants also show serological or biological divergence.

