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Evolutionary inferences from the amino acid sequence of HC-Pro, a plant virus multifunctional protein



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Short Abstract: The Helper component-proteinase (HC-Pro) encoded by viruses within the Potyviridae family is a multifunctional protein involved in several steps of the virus life cycle. We report an extensive phylogenetic analysis using the amino acid sequences of HC-Pro and provide evolutionary inferences concerning virus transmission and host range.

Long Abstract:

Potyvirus are ssRNA(+) viruses belonging to the family Potyviridae. The virus genome has a coding capacity for a single polyprotein that is proteolytically cleaved into the different active viral proteins. Among them, the nonstructural Helper-component proteinase (HC-Pro) is engaged in several steps of the potyviral life cycle, playing critical functions that were pointed out in a great number of studies. It was first described as an essential factor for virus transmission by aphid vectors. In addition to its role in transmission, HC-Pro functions as a protease catalyzing an autocatalytic cleavage at its carboxyl-terminus. Moreover, HC-Pro plays a central role in potyvirus virulence affecting genome amplification and movement and acting as a suppressor of host defense mechanisms based on RNA silencing. HC-Pro is a multifunctional protein that contains three intrinsic functional domains: an N-terminal domain essential for aphid transmission, a central domain that binds nucleic acid in a nonspecific manner and is implicated in a variety of processes, including suppression of RNA silencing and a C-terminal domain that is reminiscent of cysteine-peptidases. All sequenced viruses within the Potyviridae, including members of the genera Tritimovirus, Rymovirus, Ipomovirus and Bymovirus, encode HC-Pro homologues. However, the encoded proteins lack the specific motifs required for aphid transmission as described for potyviruses.

Searches in public databases retrieved a large number of sequences with homology to PVY HC-Pro derived from five genera of Potyviridae: Potyvirus, Rymovirus, Ipomovirus, Tritimovirus and Bymovirus. At the time of this analysis, however, no HC-Pro sequences were available for viruses within the genus Macluravirus. Sequence alignments revealed that only the C-terminal region of HC-Pro was conserved among these five genera. Although very similar among members of the Potyvirus and Rymovirus genera, the N-terminal and central regions of the protein showed a high level of variability. Taking these findings into consideration, we opted to generate two sets of multiple alignments of the HC-Pro deduced amino acid sequences; one using the C-terminal conserved region of all genera of Potyviridae and a second based on full length HC-Pro sequences of the genus Potyvirus using a rymovirus as out group.

To correlate the evolutionary history of the Potyviridae with transmission features, data concerning the vector taxa of each genus was compared with its phylogenetic position in the C-terminal-based tree. Tree topology showed the genus Bymovirus, encompassing fungi-transmitted viruses, as the most ancient of the Potyviridae and a possible ancestor of all genera. The genus Tritimovirus was placed as the second most ancient within this family.

Tritimoviruses are mite-transmitted as well as the members of the genus Rymovirus. The genus Ipomovirus, encompassing whitefly-transmitted viruses, was placed between these two mite-transmitted genera. Moreover, the economically important aphid-transmitted species of the genus Potyvirus were clustered together and showed a close phylogenetic relationship to members of the genus Rymovirus.

Species of the genera Rymovirus, Tritimovirus and Bymovirus have a narrow host range and infect only plants within the Poaceae. In contrast, species of the Potyvirus and Ipomovirus genera infect plants within the Liliopsida and Eudicotyledons. Notably, the tree based on the entire HC-Pro sequence from various potyvirus species showed a group composed of JGMV, MDMV, SrMV and SCMV that was restricted to plants within the Poaceae. These species probably represent the most ancient group within the Potyvirus and are referred to here as the Poaceae Potyvirus. Although the natural host range for other Potyvirus species is more diversified, two additional groups were identified: the Fabaceae Potyvirus (CABMV, BCMV and SMV) and the Solanaceae Potyvirus (PVY, PepMoV, PVV, WPMV and PeTMV). In general, the majority of the potyviruses show host ranges restricted to four families or less, although certain species are able to infect plants within several families: TEV (8 families), PPV (9 families) and TuMV (11 families). The most common hosts are solanaceous species followed by species within the Fabaceae and Amaranthaceae, respectively.

The phylogenetic tree indicates that HC-Pro might have originated from a Bymovirus ancestral group using members of the Poaceae family as host plants. Considering that only the C-terminal portion of HC-Pro is conserved in all members of the Potyviridae, this protein is likely to have originated as a protease. During the evolution and diversification of the different genera, the protein suffered several mutation and recombination events that resulted in a size increase and in the acquisition of new domains and functions. These newly acquired functions ended up being of great importance, such as vector transmission, cell-to-cell and long distance movement, replication and RNA silencing suppression. The fact that HC-Pro plays an essential role in aphid and mite transmission of Potyviruses and Rymovirus, respectively, indicates an important role for this protein in the evolution and diversification of the Potyviridae family. To date, however, no direct evidence in the literature confirming a role for HC-Pro in the transmission of Bymovirus by fungal-like Plasmodiophoridae exists. Nevertheless, we can reasonably deduce that HC-Pro has been fundamental for virus transmission by vectors, at least in a more recent era. The transmission mode is an important constraint of virus evolution and the acquisition of new and more efficient vectors can have a decisive impact in the appearance of new virus lineages. Thus, coevolution between virus and vector greatly influence the long-term evolutionary history of virus families. In the case of the Potyviridae family, this became evident when comparing the mobility and dispersion capacities of vectors, such as fungal-like Plasmodiophoridae and mites with that of whiteflies and aphids. By uniting the data concerning HC-Pro domain organization, HC-Pro phylogeny, host range and vectors, notable correlations were observed. When considering the facet host range x vectors, there is circumstantial evidence to suggest that only after the acquisition of more efficient vectors, like aphids, did the Potyvirus became capable of infecting a more diversified host range, therefore no longer being restricted to members of the Poaceae family.