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Potyvirus Isolation and RNA Extraction

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1. Introduction

The *Potyviridae* are the largest single group of plant viruses, and as such are the most important from an economic standpoint. There are nearly 200 distinct recognized species or other viruses that are possible or probable members of the group. Two recent books provide an excellent and in depth review of the *Potyviridae* ***(1,2)***. Generally, they are filamentous particles of ca. 11–12 × 680–900 nm. The single-stranded, message sense RNA molecule of the virus genome is encapsidated by single species of CP of ca. 30–36 kDa. This RNA (ca. 8500–9800 nucleotides) is polyadenylated and also contains a VPg covalently bound to the 5' end. Coat protein amino acid sequences show significant homology, particularly within the core region. Virions are approx 5% RNA and 95% protein by weight. The virus RNA is translated initially as a large polyprotein that is autoproteolytically cleaved to provide the mature viral proteins (**Fig. 1**).

The sheer size and diversity of this group, as well as diversity and range of susceptible hosts, makes description of general procedures difficult, at best. There are probably as many variations on the methods presented below as there are potyviruses. Some of the problems associated with purification are ameliorated by virtue of the ease with which many potyviruses can be manipulated, e.g., mechanical transmission. Although many members of the group have relatively restricted host ranges, most are not so restrictive that suitable propagation hosts cannot be found. Therefore, many of the common guidelines outlined in this chapter are applicable in terms of propagation host selection and choice of a purification procedure, if there is no precedent in the literature. Nevertheless, no single method is suitable for purification of all (or even the majority) of potyviruses.

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