

Procedures for Plant Rhabdovirus Purification, Polyribosome Isolation, and Replicase Extraction

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1. Introduction

The *Rhabdoviridae* family consists of a large number of nonsegmented negative-strand RNA viruses. As a group, the rhabdoviruses cause many serious plant and animal diseases that have detrimental effects on agricultural productivity, public health, and wildlife populations. The members of the family collectively have an unusually broad host range composed of viruses that infect both plants and animals (1). Many of these viruses are persistently transmitted to their mammalian and plant hosts by insect or arthropod vectors in which they are able to multiply. Consequently, these members of the family may have been able to expand their evolutionary diversity by use of their vectors as intermediate hosts to bridge the boundaries between the animal and plant taxa. Other rhabdoviruses are known to infect fish and aquatic invertebrates, and probably are transmitted via contaminated water.

Rhabdovirus particles are recognized easily in plants by electron microscopic observation of sap from diseased tissue or in thin sections of infected cells (2). The virions are normally bacilliform if extracts are fixed in glutaraldehyde prior to negative staining, but are bullet shaped if the fixative is omitted. Because the particles, with sizes reported to range from 45 to 100 nm wide and 150 to 400 nm long, can be distinguished so readily from the constituents present in uninfected tissue, numerous possible rhabdovirus diseases have been described in many different plant families (2). Microscopy of thin sections of infected cells reveals that the particles of different members normally have two characteristic patterns of accumulation: they are found either in association with the nucleus or in the cytoplasm. The *Sixth Report of the International Committee on Taxonomy of Viruses* (3) has used these subcellular distribution