Infectivity of Potato Spindle Tuber Viroid in Potato Plant Parts

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ABSTRACT


The infectivity of potato spindle tuber viroid (PSTV) was followed at weekly intervals up to 14 weeks after inoculation into potato (Solanum tuberosum 'Netted Gem'). The viroid was assayed on Scoporia sinensis leaves from extracts of roots, lower and upper stems, and middle and apical leaves of potato plants. The PSTV was detected from the middle leaves and roots as early as 1 week after inoculation, although infectivity was very low. The highest infectivity in most plant parts was reached 4-8 weeks after inoculation and then declined sharply. The infectivity was highest in extracts from the mature leaves in the middle of the plant and lowest in roots and tuber extracts.

Additional key words: virus-free potatoes, infectious ribonucleic acid, indexing.

Often, virus infectivity in a source plant reaches a peak 1 week after inoculation and then falls; it varies greatly with different virus-host combinations, but commonly peak infectivity corresponds with the appearance of symptoms (13). The spindle tuber disease of potatoes (Solanum tuberosum L.) is caused by a viroid (1, 9) and various soil and environmental factors that influence viroid symptom expression have been studied extensively in potatoes (3, 12). When potato spindle tuber viroid (PSTV) infectivity was studied in tomato (Lycopersicon esculentum Mill.) plants, a high infectivity was observed for a few days after the first appearance of symptoms, followed by a decrease throughout the disease development (5). There is no such information for PSTV-potato combinations. This would be useful for indexing potato plants suspected of PSTV infection. The objective of this study, therefore, was to follow the infectivity of PSTV in various parts of potato plants during disease development and estimate the suitability of those parts for PSTV indexing.

MATERIALS AND METHODS

A severe strain of PSTV that had been maintained in Scoparia sinensis Hemsl. (7) plants since 1971 was used for inoculum. The inoculum was in the form of partially purified low-molecular-weight ribonucleic acid as described earlier (9). It was used at the concentration 0.5 mg/ml in 0.1 M KH₂PO₄ buffer, pH 7.0, with a mortar and pestle. A drop of this inoculum (50 µl) was used to inoculate each half-leaf of S. sinensis plants with a glass spatula, and each sample was inoculated to two top half-leaves of five uniformly sized plants. Only one leaf per plant was used. All plant-part samples were tested within 5 days on one batch of S. sinensis seedlings grown under identical conditions of light and temperature. After inoculation, the S. sinensis seedlings were maintained as described earlier (8). The resulting local lesions were counted 15 days after inoculation.

RESULTS

Symptoms of potato spindle tuber viroid in potato cultivar Netted Gem.—When potato cuttings obtained from disease-indexed virus-free stock of cultivar Netted...
Gem were inoculated with PSTV nucleic acid and maintained under the environmental conditions described above, very distinct symptoms appeared within 3-4 weeks after inoculation. Severe stunting of the entire plant, severely reduced apical leaves, and numerous axillary shoots were evident by the 4th week. Later on, leaves became pale dull in color and necrotic streaks appeared on the petioles and the main stems. The symptoms in the disease-indexed Netted Gem cuttings were similar to those observed previously in virus-free plants. Therefore, they appeared suitable for such infection studies with severe PSTV in the greenhouse.

The relative infectivity of potato spindle tuber viroid during current year of infection.—The number of local lesions produced by the 14 weekly samples of roots, lower stems, upper stems, middle leaves, and apical leaves are presented graphically in Fig. 1. The PSTV was detected on S. sinensis as early as 1 week after inoculation in samples taken from roots and middle leaves, but infectivity was very low for the first 3 weeks. Thereafter, the infectivity increased rapidly in the middle leaves, reached a peak 7 weeks after inoculation, and then declined sharply. The next highest infectivity occurred in the upper stem, it peaked at the end of the 5th week and then declined. No PSTV was detected in apical leaves until the end of the 3rd week and even then the infectivity remained only 30% of that in the middle leaves. The peak infectivity in the apical leaves was reached 5 to 6 weeks after inoculation. Although PSTV was detected in the roots 1 week after infection and peaked at 5-7 weeks, its pattern was erratic. In the lower stems the PSTV infectivity remained low throughout the growing season, with very little fluctuation.

Infectivity comparison of different plant parts for the total period.—In order to compare plant parts on the basis of the total growth period, samples of each plant part representing 1-14 weeks were removed from storage and were combined. The combined samples of each plant part were thoroughly milled in a Wiley mill (Arthur H. Thomas, Laboratory Apparatus, Philadelphia) fitted with a 0.5-mm (40-mesh) screen. About 300 mg of milled samples of each plant part were homogenized in 6 ml of 0.1 M K$_2$HPO$_4$ buffer, pH 7.0, and each sample was inoculated to 20 half-leaves. A tuber sample also was included in this experiment. A 3-gram sample of fresh tuber tissue was taken from each of six different tubers and combined before being freeze-dried as the other samples had been. The middle leaves and the upper stem contained the highest infectivity on the basis of the whole growing season. The lower stem and apical leaves had about half the infectivity of the leaf from the middle of the plant or upper stem, and the roots about half again as much. The tubers had less than 10% of the viroid contained in the middle leaves.

Comparison of relative infectivity from tissue extract vs. nucleic acid.—The above experiment shows that differences in PSTV infectivity from various plant parts may be due to their differential release from the tissues, or to varying content of ribonucleases; it may not reflect the true PSTV content of the plant parts. To eliminate these possibilities, the following tests were made: 2 g of freeze-dried tissue representing each plant part was removed

![Graph of relative infectivity](image)

**Fig. 1.** The infectivity of potato spindle tuber viroid in different parts of potato plants at various periods after inoculation.
TABLE 1. Comparison of infectivity of tissue extract and nucleic acid preparations from various potato plant parts of potato spindle tuber viroid-infected plants of potato cultivar Netted Gem

<table>
<thead>
<tr>
<th>Plant parts</th>
<th>Tissue extract</th>
<th>Nucleic acid</th>
</tr>
</thead>
<tbody>
<tr>
<td>Apical leaves</td>
<td>25</td>
<td>22</td>
</tr>
<tr>
<td>Upper stem</td>
<td>64</td>
<td>55</td>
</tr>
<tr>
<td>Middle leaves</td>
<td>65</td>
<td>92</td>
</tr>
<tr>
<td>Lower stem</td>
<td>34</td>
<td>25</td>
</tr>
<tr>
<td>Root</td>
<td>16</td>
<td>30</td>
</tr>
</tbody>
</table>

*Average of 10 half-leaves of Scopolia sinensis; each tissue extract and respective nucleic acids were paired on each plant and they occurred on left and right half-leaves five times. Only two half-leaves were inoculated on each plant.

The differences in the infectivity of various parts probably reflect viroid content of the tissues involved and not just extraction facility, because both 'extract' or 'nucleic acids' from each part produced a similar number of local lesions.

The pattern of infectivity fluctuation in most plant parts appears typical of any virus-host combination in that the infectivity peaked when symptom development was most severe and declined thereafter. In this regard the PSTV-potato combination is similar to the PSTV-tomato combination studied earlier (5).

**LITERATURE CITED**