RELATIVE PREVALENCE OF MILD AND SEVERE STRAINS OF POTATO SPINDLE TUBER VIRUS IN EASTERN CANADA

R. P. SINGH, R. E. FINNIE AND R. H. BAGNALL[1]

ABSTRACT

In 1969, 355 leaf or tuber samples from potatoes suspected of harboring the potato spindle tuber virus were collected from 23 tablestock fields in eastern Canada. By means of Fernow's tomato cross protection test, modified by the use of ribonucleic acid extract as inoculum, 317 samples were shown to be infected, 290 (92%) of these with a mild strain, and 27 (8%) with a severe strain of the virus. Preliminary tests in the greenhouse and field indicate that the mild strain, so named because of symptoms on tomato, is relatively mild in the potato as well.

RESUMEN

En 1969, 355 muestras de hojas o tubérculos posiblemente infectados con el virus de tubérculos ahusados (PSTV) fueron collectados de 23 campos comerciales en el Este del Canadá. Por media del ensayo de proteción cruzad en tomate de Fernow, modificado con el uso de extracto del ácido ribonucleico como inóculo, 317 muestras fueron probados como infectados, 290 (92%) con la blanda, y 27 (8%) con la cepa virulenta del virus. Ensayos preliminares en el invernadero y campo indican que la cepa blanda, llamado así debido a los sintomas en tomate, es también relativamente blanda en la papa.

When the tomato was advanced as a test plant for the potato spindle tuber virus (PSTV) (4, 7), it soon became clear that there existed a strain or strains of the virus that would not cause diagnostic symptoms in this host. Fernow (1) developed a technique for detecting these mild strains (MPSTV), based on cross-protection in tomato against the severe or "Schultz" strain (SPSTV). Subsequenlty, Fernow, Peterson, and Plaisted (2) found that a comprehensive system of field and harvest inspections and greenhouse eye-indexing, supplemented by their tomato cross-protection test would keep PSTV infections in their field plots to a very low level. But individual inspections and tomato tests were still not entirely consistent. This was particularly so where inoculum for the tomato test was taken directly from tubers in the fall after harvest — an ideal time to commence testing if an extensive program is planned.

We have found ribonucleic acid (RNA) extract from SPSTV material to be more infectious than raw tuber or foliage juice (5, 6), but the extraction procedure was too involved for use in a diagnostic testing program. Here, we have used a simplified RNA extraction procedure combined with Fernow's tomato cross-protection test in an attempt to improve the efficiency of the latter for detection of both SPSTV and MPSTV. The RNA extraction has an added advantage in that potato viruses X and Y are eliminated. These viruses sometimes complicate the symptom picture in tomato.

[1]Canada Department of Agriculture, Research Station, Fredericton, New Brunswick, Canada. Accepted for publication December 22, 1969.

Further, Fernow and his co-workers (1, 2) observed that the mild strain predominated in their field plots at Ithaca. We have had similar experience at Fredericton and have extended the search through commercial potato stocks in four provinces of eastern Canada. Some yield comparisons between healthy, SPSTV- and MPSTV-infected potatoes were also made.

Materials and Methods

The RNA extract was prepared by grinding at room temperature, approximately 6 g of foliage or tuber tissue in a mortar containing 4 ml of water-saturated phenol. (To avoid splashing the caustic phenol, the tuber sample was precrushed, phenol added, and crushed further). The slurry was centrifuged and the aqueous top layer (2 to 3 ml) containing RNA, decanted, washed with about 2 volumes of ether to remove the residual phenol, and centrifuged again. Here, the aqueous layer still containing the RNA, was drawn off from the bottom, and if not used within a day or so for inoculation, it was frozen.

To diagnose MPSTV, we used the method of Fernow et al whereby two plants of Sheyenne tomato were inoculated at the cotyledon stage with our RNA extract. Fourteen days later, if there was no evidence of SPSTV symptoms (5, 7), one of the two plants was inoculated a second time or 'challenged' with SPSTV-RNA. Three weeks later, the presence of 'severe' symptoms on the challenged plant was interpreted to mean that it had been uninfected at the time of the challenge. Conversely, if no symptoms resulted from the challenge, cross-protection by MPSTV was assumed. Meanwhile, the second plant, inoculated only once, was held for observation of possible late-developing symptoms of SPSTV or for slight signs of MPSTV. For checks, RNA extracts from healthy potato plants — our RNA extract includes most of the plant RNA's as well as any PSTV-RNA — were used in the initial inoculation. When challenged with SPSTV-RNA, severe symptoms almost invariably resulted.

Experiment and Discussion

From an experimental plot at Fredericton, 132 hills of Netted Gem potato were selected for the cross-protection test. On the basis of foliage symptoms, 117 were identified as infected with PSTV and the remaining 15 as 'healthy'. Foliage RNA was used to inoculate the tomato test plants. All of the plants with foliage symptoms were found to be infected, two with SPSTV and 115 with MPSTV. Six of the 'healthy' lot were found by means of the challenge test to be infected with MPSTV — possibly too recently for symptom development — and the remaining nine gave no evidence of infection. At harvest, tubers from each hill were examined for evidence of the disease. Only 107 of the 117 plants with foliage symptoms and none of the 15 'healthy' plants produced spindled tubers. The cross-protection test with RNA inoculum appeared to be quite effective in the detection of MPSTV, and in the field, at least under our conditions, diagnosis by means of foliage symptoms was superior to diagnosis based on tuber symptoms.

Next, we made a preliminary survey to determine the distribution of the different strains of PSTV in Eastern Canada. Samples of different varieties were obtained from four provinces (Table 1). The samples were

TABLE 1.—*Mild and severe strains of potato spindle tuber virus in foliage and tuber samples selected from tablestock fields in eastern Canada.*

Province	Variety	No. of fields	Recovery of PSTV None	Severe	Mild
Quebec[1]	Kennebec	5	11	5	33
	Keswick	2	0	2	8
New Brunswick[2]	Kennebec	4	12	10	29
	Netted Gem	5	13	4	167
Prince Edward Island[2]	Kennebec	2	0	2	12
	Netted Gem	2	0	1	13
	Sebago	2	0	2	19
Newfoundland[1]	Pink Pearl	1	2	1	9
Total		23	38	27	290

[1]Inoculum prepared from tubers.
[2]Inoculum prepared from foliage.

selected mainly from fields planted for table stock where diseased plants were most readily found. Tuber samples collected by potato inspectors were obtained from Quebec and Newfoundland, whereas tuber and foliage samples were collected by ourselves in New Brunswick and Prince Edward Island.

Of the 355 samples from 23 fields, 317 (89%) were infected, as determined by the RNA-challenge test. Of those infected, 290 (92%) had MPSTV and 27 (8%) SPSTV. This confirms in commercial plantings, the observation by Fernow (1) and ourselves that the mild strain predominated in experimental field plots.

Since all selections were made in belief that the plants or tubers were probably infected, it is intersting to note that 23 of the 38 samples (60%) that proved to be free of PSTV were Kennebec, whereas this variety represented 114 in the total of 355 samples (only 32%). Evidently, it was difficult to diagnose PSTV in this variety, and several potato inspectors have agreed with this observation. A further observation on which we can only speculate at the moment, is that all 49 samples selected on Prince Edward Island were infected, although the balance between MPSTV and SPSTV was quite similar to that obtained elsewhere.

Effect of MPSTV on yield

Our data on the effect of MPSTV on yield comes from two sources:

(a) *Greenhouse yield trial:* Preliminary to the actual trial four lots (three plants each) of healthy plants of the variety Saco were grafted with tomato scions infected, respectively, with (i) and (ii) MPSTV from two different sources supplied by Dr. K. Fernow, (iii) MPSTV from a Kennebec tuber obtained on Prince Edward Island, and (iv) SPSTV from a Saco tuber obtained from the Schultz Virus Collection (8). All of the grafted Saco produced spindle-shaped tubers, and these plus tubers from ungrafted healthy Saco were used to plant the yield trial. This trial consisted of six replicates of each of the four infected lots plus the healthy check; each replicate consisting of five plants grown in

7-inch clay pots. While 30 plants from each lot were required in the trial, these were actually selected soon after emergence from 50 that had been planted. Thus the plants in all five lots were approximately equal at this stage. The healthy lot produced a total of 3837 g of tubers, the two Fernow MPSTV-infected lots 3645 g (5% reduction) and 3401 g (11%), the Kennebec MPSTV-infected lot 3100 g (19%), and the SPSTV-infected lot 2071 g (46%), respectively. This trial was intended as a multiplication step toward a future field trial, but the results are sufficiently interesting to be mentioned here. A significant feature in this greenhouse trial was that practically all plants infected with MPSTV assumed the classic symptoms — upright growth habit with sharply angled petioles. This contrasted with the gracefully draping leaves on the healthy plants. The plants infected with SPSTV also had the upright growth habit, but were stunted and somewhat distorted, with necrotic lesions on the stems and petioles — symptoms seldom seen in 'spindle tuber' diseased specimens in commercial fields, and clearly not the normal picture of the disease familiar to potato inspectors.

(b) *Field selection and yield:* In a half-acre field of Green Mountain potatoes grown in 5-set tuber units, about one-third of the units were observed to have upright growth habit and the remainder a normal spreading growth. Upon testing by means of the tomato challenge test, it was determined that plants in the 'upright' units were infected with MPSTV. Plants in the 'normal' units — early in the season at least — were healthy. On September 9 (planted on May 28) ten units of normal and ten units of MPSTV plants, selected at random, were harvested. The 'normal' plants produced an average of 1156 g of tubers per plant, while the MPSTV plants produced 988 g (15% reduction). On September 15, another ten units of each lot were harvested. The 'normal' plants produced 1372 g of tubers per plant against 1098 g (20% reduction) for the MPSTV plants. Yield reduction of this order is important yet modest when compared with 65% reported by Hunter and Rich (3) for the SPSTV strain.

Our yield tests are preliminary, but they serve to show that MPSTV isolates are mild in potatoes as well as in tomatoes, relative to the SPSTV strain. Nevertheless, these mild isolates do cause misshapen tubers, and reduce yield. There is some suggestion in the results from the greenhouse trial that isolates of the MPSTV differ from each other and it seems possible that strains so mild as to be termed 'latent' might exist.

Literature Cited

1. Fernow, K. H. 1967. Tomato as a test plant for detecting mild strains of potato spindle tuber virus. Phytopathology 57: 1347-1352.
2. Fernow, K. H., L. C. Peterson and R. L. Plaisted. 1969. The tomato test for eliminating spindlle tuber from potato planting stock. Amer. Potato J. 46: 424-429.
3. Huunter, J. E. and A. E. Rich. 1964. The effect of potato spindle tuber virus on growth and yield of Saco potatoes. Amer. Potato J. 41: 113-116.
4. Raymer, W. B. and Muriel J. O'Brien. 1962. Transmission of potato spindle tuber virus to tomato. Amer. Potato J. 39: 401-408.
5. Singh, R. P. and R. H. Bagnall. 1968. Infectious nucleic acid from host tissues infected with the potato spindle tuber virus. Phytopathology 58: 696-699.

6. Singh, R. P. and R. H. Bagnall. 1968. *Solanum rostratum* Dunal. a new test plant for the potato spindle tuber virus. Amer. Potato J. 45: 335-336.
7. Singh, R. P., A. P. Benson and F. M. Salama. 1964. Sheyenne tomato variety as an indicator for potato spindle tuber virus. (Abstr.). Amer. Potato J. 41: 304.
8. Webb, R. E. 1958. Schultz potato virus collection. Amer. Potato J. 35: 615-619.