Resistance to Beet Western Yellows Virus Among Forage Brassicas

P. E. THOMAS, Research Plant Pathologist, USDA-ARS, and D. W. EVANS, Agronomist, Washington State University, Irrigated Agriculture Research and Extension Center, Prosser 99350-9687; and L. FOX, Biological Technician, and K. D. BIEVER, Research Entomologist, USDA-ARS, Yakima Agricultural Research Laboratory, Yakima, WA 98902

ABSTRACT

Thomas, P. E., Evans, D. W., Fox, L., and Biever, K. D. 1990. Resistance to beet western yellows virus among forage brassicas. Plant Dis. 74:327-330.

Fourteen forage *Brassica* cultivars representing four species were evaluated for resistance to beet western yellows virus (BWYV), potato leafroll virus (PLRV), and the tomato yellow top strain of PLRV (PLRV-TY) under field exposure. Incidence of field infection with BWYV ranged in several susceptibility classes from less than 2 to 100%. The four most susceptible cultivars were all turnips. The four most resistant cultivars included one each of the four *Brassica* species. All four species were represented among the intermediately susceptible cultivars. Once infection was achieved, there was little or no difference among the cultivars in resistance to systemic virus accumulation. Symptoms of BWYV were mild. Neither strain of PLRV was detected by ELISA or aphid transmission. There was no correlation between numbers of aphids on plants in the fall and susceptibility to infection. All *Brassica* cultivars survived winters, but neither live aphids (*Myzus persicae*) nor aphid eggs were found on plants at the end of winter.

In the intermountain regions of the northwestern United States and in many other areas across the nation, there is a growing interest in the use of fall-seeded *Brassica* cultivars as winter forage crops. Seeded as a second crop in July and August, following early potatoes, sweet corn, peas, cereal grains, and other primary crops, the brassicas take advantage of open land that otherwise remains idle until the next spring under current cropping systems. The achievable yields are high (9,10,14) and the potential for expansion of the crop is great. In Washington alone, some 80,000 open, irrigated hectares are available (24).

Cooperative investigations of the USDA-ARS and the Washington State University Agricultural Research Center, Prosser. Plant Pathology PPNS 0045, College of Agriculture and Home Economics Research Center, Pullman, WA 99164.

Accepted for publication 7 November 1989 (submitted for electronic processing).

Brassica species are also spring-seeded in many areas and used during summer, fall, and winter as forage.

Various *Brassica* species are known to be primary hosts of beet western yellows virus (BWYV) (3,4), and large populations of its primary vector, the green peach aphid (*Myzus persicae* (Sulzer)), are known to occur in the fall in the Northwest (17,18,21) at a time when the fall-seeded *Brassica* crop is beginning to grow. Plants are normally most susceptible to virus infection and most severely affected by virus disease when young (1). Thus, it seemed probable that forage brassicas grown as a second, fall-seeded crop in the Northwest could be exposed to high inoculum pressures to BWYV and could vary in their field susceptibility to this virus.

Because many *Brassica* species survive winters in the Northwest, fall-seeded brassica forage crops could significantly impact on the epidemiology of a number of viral diseases in other crops by serving as an overwintering source of virus inoculum. Of particular interest are potatoes, a major crop of the Northwest, and spring seed rape, which is rapidly becoming an important cash crop there. BWYV was reported to be involved in the potato leafroll syndrome (6–8), a disease that reduces yield and quality of tubers (11,23). It is also known to infect and to significantly reduce vigor and yield of seed rapes (5,12,20).

Because potato leafroll virus (PLRV) was isolated from some *Brassica* winter annual weeds in Washington (22), it seemed possible that PLRV or its tomato yellow top strain (PLRV-TY) (13) could overwinter in *Brassica* cultivars. Both viruses were transmitted to *Capsella bursa-pastoris* L. (13) but could not be transmitted to *B. chinensis* L., to *B. nigra* (L.) Koch, or to three cultivars of *B. oleracea* L. (13). Although Salaman and Wortley (19) transmitted PLRV to *Matthiola*, turnip, and brussels sprouts, Helson and Norris (15) failed to transmit PLRV to these or to other crucifers.

As a part of the evaluation of *Brassica* cultivars for potential use as forage crops in the Northwest, the purpose of this study was to determine the field susceptibility of *Brassica* cultivars to BWYV, PLRV, and PLRV-TY and to determine the potential impact of producing forage *Brassica* crops on the epidemiology of other virus diseases.

MATERIALS AND METHODS

The incidence of field infection among the *Brassica* cultivars was used as a measure of resistance to infection, i.e., ability to suppress initial virus infection. Virus-specific serological reactivity of triturated tissue extracts was used to measure ability of plants to suppress systemic virus accumulation following infection. The resistance to infection and to systemic virus accumulation by BWYV, PLRV, and PLRV-TY was compared in 14 forage *Brassica* cultivars in field plots in 1986 and 1987. The cultivars included four forage rapes (*B. napus* L. 'Ramon,' 'Windal,' 'Velox,' and 'Jumbo'), two swedes (*B. napobrassica* L. 'Seefelter' and 'Angela'), six turnips (*B. rapa* L. 'Rondo,' 'Mosa,' 'Goldvital,' 'Purple Top' [White Globe], 'Forage Star' [hybrid], and 'Barive' [cow turnip]), and two kales (*B. oleracea* var. *viridis* L. 'Premier' and 'Maris Kestral').

The cultivars were seeded in a randomized complete block design with three replicates in the first year (1986) and five in the second (1987). Plots were 1.4 × 6.6 m, with 0.3 m between plots, the first year and 1 × 3 m the second year. Row spacing was 15 cm. Seeding dates were 8 August 1986 and 17 August 1987. The plots were arranged side by side across a field. In 1986, the virus plots represented one-half of a larger set of plots; the other half was used for agronomic evaluations. Thus, each plot was bordered on one side by an equal-sized plot of the same *Brassica* cultivar and on the other by an alfalfa field. In 1987, the plots were next to a larger field of Purple Top turnips (seeded 15 days earlier) on one side and to a freshly seeded wheat field on the other. Three potato plots (about 8 ha) used to provide severe exposure of potato breeding progeny to PLRV were within 400 m of the *Brassica* plots.

The *Brassica* plantings followed oats (*Avena sativa* L.) in the rotation both years. The soil, a silt loam, tested each year above P & K response levels. Ammonium nitrate was broadcast on the growing crop at rates of 134 kg of nitrogen per hectare on 2 September 1986 (year 1) and 150 kg of nitrogen per hectare on 20 August 1987 (year 2). To control weeds, trifluralin (Treflan 4E) was preplant-incorporated to 10 cm at rates of 0.42 kg a.i./ha in year 1 and 0.93 kg a.i./ha in year 2. The plots were sprinkler-irrigated at 7- to 14-day intervals.

Plants were assayed for virus content using a two-step adaptation (16) of the standard double antibody sandwich enzyme-linked immunosorbent assay (ELISA) (2). All antisera used were prepared in rabbit in our laboratory. The BWYV antiserum was prepared against a BWYV isolate supplied by J. E. Duffus, USDA-ARS, Salinas, California, which was originally isolated from radish (*Raphanus sativus* L.) and designated RY1R. The PLRV antiserum was prepared against an isolate designated as LR-7, which produces severe net necrosis leafroll symptoms in Russet Burbank and other potatoes susceptible to net necrosis. It was isolated in the Yakima Valley of Washington and supplied by Lee Fox, USDA-ARS, Yakima, Washington. The PLRV-TY antiserum was prepared against an isolate from Prosser, Washington, designated YT-79.

The same γ-globulin and conjugate preparations were used at the same dilutions and under the same conditions in all assays. Final γ-globulin dilutions used to coat plates were 1:1,000, 1:5,000, and 1:2,000 for BWYV, PLRV, and PLRV-TY, respectively; corresponding conjugate dilutions were 1:4,000, 1:10,000, and 1:800. ELISA samples were prepared from four 3-mm-diameter leaf disks from each of three young leaves of each plant (fresh weight about 30 mg). The leaf disks were combined and triturated in 0.1 ml of ELISA buffer in a conical-shaped, 1.5-ml polyethylene centrifuge tube with a spinning Teflon pestle. An additional 0.5 ml of ELISA buffer was added to each tube and mixed, and 0.1 ml of the final tissue macerate was added to 0.1 ml of conjugate in an ELISA plate well precoated with γ-globulin. The ELISA plates were incubated in a moist chamber at 4 C for 18 hr, then washed. *p*-Nitrophenyl phosphate substrate solution was added, and A_{405} in each well was read spectrophotometrically after 1 hr at 23 C using a Bio-Tek model EL 307 EIA reader. The reader was zeroed on buffer control wells.

The sample dilution used in the ELISA assays (about 1:40) was selected from the midpoint of the linear portion of a sample dilution series. This was done to ensure that differences in antigen concentration among the infected plants would be reflected as differences in the A_{405} values obtained in the assays. The dilution series was prepared from a composite of tissue taken from three infected Purple Top turnip plants growing in the experimental plots.

Both healthy and infected control samples were prepared from greenhouse-grown *Physalis floridana* L. plants. To ensure that healthy and infected control samples remained constant in all assays each year, a bulk sample was prepared as described for test samples, divided into aliquots, and held at −20 C. Aliquots were removed from the freezer and used as needed.

Ninety plants of each cultivar (30 plants from each of three replicate plots of each cultivar in 1986 and 18 plants from each of five replicate plots in 1987) were assayed by ELISA for virus content in November each year. A single well assay from each of the 90 plants of each cultivar plus two wells with healthy control tissue, two with infected control tissue, and two with buffer controls completely filled a 96-well ELISA plate.

Aphid transmission assays were conducted on selected plants to supplement and confirm ELISA results. Nonviruliferous green peach aphids were held for about 30 hr on a young leaf of each plant to be assayed in a petri plate on moist filter paper. The aphids were then caged on index host seedlings for 48 hr under a plastic cylinder with one end covered with nylon screen. Aphids were killed by fumigation with nicotine sulfate. The index hosts (turnip cv. Goldvital and *P. floridana*) were observed for symptom development and were assayed by ELISA after 4 wk.

On 22 September in the second year of testing, the numbers of alate and apterous green peach aphids and all other aphids were counted on five leaves harvested from each of the five replicate plots. The plants were examined for overwintering live aphids and for aphid eggs at the end of February 1987.

Table 1. Resistance to infection with beet western yellows virus (BWYV) among field plants of 14 forage brassica cultivars

		Incidence of infection[x]				
		No. infected[y]		Rank		
Cultivar	Species	1986	1987	1986	1987	Percent[z]
Goldvital	Turnip	89	90	1	1	99 a
Rondo	Turnip	76	88	3	3	91 a
Mosa	Turnip	66	90	4	2	87 a
Barive	Turnip	78	NT	2	NT	87 a
Windal	Rape	47	77	6	5	69 b
Angela	Swede	48	54	5	7	57 c
Purple Top	Turnip	27	68	8	6	53 c
Jumbo	Rape	29	43	7	10	40 d
Premier	Kale	17	46	9	8	35 d
Velox	Rape	12	46	10	9	32 d
Ramon	Rape	7	29	13	11	20 e
Seefelter	Swede	11	NT	11	NT	12 ef
Forage Star	Turnip	11	10	12	13	12 ef
Maris Kestral	Kale	1	2	14	14	2 f

[x] Determined by ELISA, supplemented on selected plants by aphid transmission assays. NT = not tested.
[y] Number of plants infected among 90 assayed.
[z] Combined results for 1986 and 1987. Values followed by different letters are significantly different.

RESULTS

As reflected by incidence of infection (Table 1), the *Brassica* cultivars ranged from highly resistant to infection by BWYV to highly susceptible. Less than 2% of the most resistant cultivar, Maris Kestral, were infected, while incidence of infection approached 100% among the four most susceptible cultivars—Goldvital, Rondo, Mosa, and Barive. All Mosa and Goldvital plants assayed were infected in the second year. The 14 cultivars were divided into several distinct susceptibility categories by statistical analysis. Although the four most susceptible cultivars were all turnips, the four most resistant cultivars included one of each of the *Brassica*

species used in this study. All four species were represented among intermediately susceptible cultivars.

Incidence of infection was higher in 1987 than in 1986 (Table 1), but the relative ranking of the cultivars remained about the same. Notable exceptions were Purple Top, which was relatively more susceptible in 1987, and Jumbo, which was relatively more resistant.

To determine the incidence of infection, it was necessary first to determine the correspondence between the distribution of ELISA values obtained for the 90 plants assayed in each cultivar (Table 2) and actual infection. This was done using aphid transmission from selected plants in the field to indicator hosts in the greenhouse. In each cultivar, the first three plants with ELISA readings above absorbance 0.099, the first 10 plants (or all plants when fewer than 10 were available) with absorbance values falling below 0.050, and all plants with absorbance values falling between 0.050 and 0.100 were assayed by aphid transmission. Plants scheduled for aphid transmission assays were pruned, transplanted, and held for 4–6 wk in the greenhouse and then assayed both by aphid transmission and by ELISA.

In aphid transmission assays conducted to determine the correspondence between ELISA results and actual infection, all plants that had produced A_{405} values above 0.099 (Table 2) in the field proved to be infected. Forty-six of 100 plants that produced A_{405} values in the range of 0.050–0.099 were infected, and 22 of 227 plants that produced A_{405} values below 0.050 were infected. Although all of the plants assayed for infection by aphid transmission had produced relatively low ELISA values when sampled in the field, they produced values approximating those of control plants after they had been pruned and regrown in the greenhouse.

Although some cultivars apparently were much more difficult to infect than others in the field, there was little, if any, difference among the cultivars in capacity of virus to accumulate systemically once infection was achieved (Table 2). The modal frequency of ELISA readings was about the same for all of the cultivars. Although modal frequency was somewhat higher for the most susceptible cultivar, Goldvital, the mean A_{405} value for infected control tissue was also higher in that distribution. The distributions of the most susceptible cultivars were somewhat skewed in favor of higher values. A few plants, notably among Rondo, Windal, and Mosa, produced very high ELISA values that exceeded the reading limits of the reader.

None of the 180 plants in each cultivar assayed over 2 yr produced a clearly positive ELISA for PLRV or PLRV-TY. To test the possibility that PLRV might be present in the *Brassica* cultivars at concentrations below the detection limits of ELISA, the four plants of each cultivar in each year with the highest ELISA readings for PLRV and PLRV-TY were pruned, transplanted in the greenhouse, and later assayed by aphid transmission to *P. floridana*. None produced a positive assay for either virus 4 wk later.

Many green peach aphids were found on plants of all the *Brassica* cultivars in the field. There were no significant differences in numbers of aphids on plants of the various cultivars (Table 3), however, and there was no correlation between numbers of aphids on plants and susceptibility to initial infection. Neither live aphids nor eggs overwintered on any *Brassica* plants.

Infection with BWYV was not associated with strong visible symptoms or reductions in vigor or size of plants among any of the cultivars. Among the turnips and swedes, plants that produced positive ELISA results usually had a

Table 2. Frequency distribution of ELISA readings (A_{405}) among 90 plants in each of 14 forage brassica cultivars assayed for beet western yellows virus (BWYV)

		ELISA reading (A_{405}) categories[z] (no. of plants)								
Cultivar	Season	0–0.049	0.050–0.099	0.100–0.199	0.200–0.399	0.400–0.599	0.600–0.799	0.800–0.999	1.00–1.99	≥2.00
Goldvital	1	3	2	6	9	*26*	20	12	12	0
	2	0	2	7	11	*20*	19	13	14	4
Barive	1	15	2	6	*23*	22	11	2	9	0
	2	NT	NT	NT	NT	NT	NT	NT	NT	NT
Rondo	1	16	10	19	*15*	10	2	4	11	3
	2	3	0	3	*12*	14	11	10	20	17
Mosa	1	21	9	*15*	10	9	10	6	10	0
	2	1	2	4	*17*	22	9	10	17	8
Windal	1	52	4	*12*	8	2	5	0	3	4
	2	12	5	*10*	12	9	4	10	13	15
Angela	1	42	5	*17*	11	9	1	3	2	0
	2	40	8	*13*	18	5	3	2	1	0
Purple Top	1	68	5	*11*	5	1	0	0	0	1
	2	27	8	*19*	20	8	8	2	3	0
Jumbo	1	66	1	7	*8*	4	3	1	0	0
	2	45	6	15	*12*	8	2	2	2	0
Premier	1	68	6	*13*	1	2	0	0	0	0
	2	38	8	12	*10*	12	5	3	2	0
Velox	1	76	2	*6*	5	0	1	0	0	0
	2	45	4	*11*	17	6	4	0	3	0
Seefelter	1	78	1	*5*	3	2	0	1	0	0
	2	NT	NT	NT	NT	NT	NT	NT	NT	NT
Ramon	1	0	7	*4*	0	0	0	1	0	0
	2	60	1	*8*	13	3	1	1	3	0
Forage Star	1	80	0	*7*	2	0	1	0	0	0
	2	81	2	*6*	1	0	0	0	0	0
Maris Kestral	1	89	0	*1*	0	0	0	0	0	0
	2	87	1	*1*	0	0	1	0	0	0

[z]ELISA reader was zeroed at 0.010 absorbance units below the mean value of the healthy control plant samples. Absorbance category into which infected control plant samples fell is in italics for each cultivar. NT = not tested.

Table 3. Number of green peach aphids (*Myzus persicae*) and all other aphids on leaves of 13 forage brassica cultivars on 22 September 1987[y]

	No. of green peach aphids			No. of other aphids			Total no. of aphids[z]
Cultivar	Alate	Apterous	Total	Alate	Apterous	Total	
Goldvital	1	3	4	0	5	5	9
Rondo	0	6	6	0	2	2	8
Mosa	1	27	28	0	0	0	28
Windal	0	5	5	0	2	2	7
Angela	0	5	5	1	0	1	6
Purple Top	1	11	12	1	3	4	16
Jumbo	0	4	4	1	1	2	6
Premier	1	10	11	0	0	0	11
Velox	0	5	5	1	0	1	6
Seefelter	0	5	5	2	11	13	18
Ramon	1	16	17	1	4	5	22
Forage Star	1	12	13	0	3	3	16
Maris Kestral	0	7	7	1	8	9	16

[y]Total number of aphids on five leaves from each of five replicate plots of each cultivar.
[z]There were no significant differences among cultivars in numbers of aphids.

slight to moderate yellow cast. The kales and rapes were not visibly affected by infection.

DISCUSSION

Although it was previously known that BWYV infects various *Brassica* species (3,4) and may cause losses in seed rapes (5,12,20), we believe this is the first documentation that forage *Brassica* cultivars may be highly susceptible to field infection by BWYV and that effective field resistance to infection is available in cultivars of major *Brassica* species. Although Gilligan et al (12) suggested that differential susceptibility to BWYV could account for the expression of a disease in one seed rape cultivar but not in others, they failed to demonstrate that the disease was caused by BWYV.

These studies do not distinguish whether the suppression of field infection observed in the *Brassica* cultivars is conditioned by mechanisms operative before, during, or after introduction of virus into the plant. However, the fact that the primary insect vector of BWYV was observed colonizing all of the *Brassica* cultivars without distinction indicates that vector nonpreference is not a factor in the resistance observed.

Once plants were infected, capacity of BWYV to accumulate systemically was about the same in all cultivars, i.e., resistance to systemic virus accumulation was not associated with resistance to infection. The fact that the mode A_{405} value in the ELISA frequency distribution was somewhat higher for the most susceptible cultivar is at least partially explicable on the basis that the A_{405} values for infected control tissue were also higher in that assay. Some unknown factor apparently caused all absorbance values in the ELISA plate from which that distribution was derived to be higher. The slight skew in favor of higher values in the frequency distributions for the most susceptible cultivars could reflect an earlier infection date for cultivars with greater susceptibility to infection. An earlier infection date would provide more time for systemic virus accumulation before the sampling date.

A number of plants that initially produced low or negative ELISA readings later proved to be infected by aphid transmission assays. The fact that these plants later produced ELISA readings in the normal range after a period of incubation in the greenhouse indicates that at the time of field sampling, infection had not yet achieved full systemic development. The low content and inconsistent distribution of virus in these plants were reflected in low and inconsistent ELISA results.

The fact that several infected plants escaped detection by ELISA illustrates the necessity for determining the correspondence between actual infection and the results of any indirect assay method to determine incidence of infection in a field population. No statistical manipulation would detect the escaped plants until the correspondence between actual infection and ELISA results was known. Furthermore, it would be impossible to produce infected controls identical to test plants because time of infection of test plants could not be known.

Unfortunately, the difference in yield between infected and healthy plants was not measured. The recency of infection among the brassicas may account, at least partially, for the mild symptoms observed and the failure of disease to cause visually detectable reductions in growth. However, lack of symptoms is no certain indication that yield loss did not occur. Asymptomatic viral infections commonly cause significant reductions in yield (25), and asymptomatic BWYV infection is known to cause marked reductions in oilseed rape yields (20).

It is doubtful whether forage brassicas could ever serve as overwintering hosts for either strain of PLRV. Neither virus could be isolated from or detected in any of the field-exposed plants, and there is good reason to believe that exposure was severe. The major vector of BWYV and PLRV (green peach aphid) was abundant on the *Brassica* plants, and the susceptible *Brassica* cultivars were readily infected with the related BWYV. All susceptible potato plants grown in nearby plots without pesticide protection were infected with PLRV during the 2 yr of this study (M. W. Martin, *personal communication*), as they have routinely been in the past (21).

Since the forage brassicas survive winters in the Northwest, the susceptible cultivars could provide an excellent overwintering source for BWYV. Thus, the widespread use of BWYV-susceptible *Brassica* cultivars in the Northwest could impact diseases in crops infected by BWYV. Because neither aphid eggs nor live aphids overwintered on the *Brassica* crop, spring dissemination from that crop would be delayed until after its colonization with aphids from outside sources. Use of the BWYV-resistant cultivars identified in this study could largely eliminate any impact of the forage brassicas on the epidemiology of BWYV-induced diseases of other crops.

LITERATURE CITED

1. Bawden, F. C. 1950. Plant Viruses and Virus Diseases. Chronica Botanica Co., Waltham, MA. 335 pp.
2. Clark, M. F., and Adams, A. N. 1977. Characteristics of the microplate method of enzyme-linked immunosorbent assay for the detection of plant viruses. J. Gen. Virol. 34:475-483.
3. Duffus, J. E. 1960. Radish yellows. A disease of radish, sugar beet, and other crops. Phytopathology 50:389-394.
4. Duffus, J. E. 1964. Host relationships of beet western yellows virus strains. Phytopathology 54:736-738.
5. Duffus, J. E. 1975. Effect of beet western yellows virus on crambe in the greenhouse. Plant Dis. Rep. 59:886-888.
6. Duffus, J. E. 1981. Distribution of beet western yellows virus in potatoes affected by potato leaf roll. Plant Dis. 65:819-820.
7. Duffus, J. E. 1981. Beet western yellows virus isolated from potatoes with the leaf roll syndrome. (Abstr.) Phytopathology 71:105.
8. Duffus. J. E. 1981. Beet western yellows virus—a major component of some potato leafroll-affected plants. Phytopathology 71:193-196.
9. Evans, D. W., Bower, D. B., and Cline, T. A. 1988. Species and planting dates for second crop forage production. Wash. Agric. Exp. Stn. Res. Bull. 0996.
10. Evans, D. W., and Cline, T. A. 1988. Second crop purple top turnip: Population, yield, and yield distribution. Wash. Agric. Exp. Stn. Res. Bull. XB0990.
11. Gilbert, A. H. 1928. Net necrosis of Irish potato tubers. Vermont Agric. Exp. Stn. Bull. 289. 36 pp.
12. Gilligan, C. A., Pechan, P. M., Day, R., and Hill, S. A. 1980. Beet western yellows virus on oilseed rape. Plant Pathol. 29:53.
13. Hassan, S., Thomas, P. E., and Mink, G. I. 1985. Tomato yellow top virus: Host range, symptomatology, transmission, and variability. Phytopathology 75:287-291.
14. Heineman, W. W., Hinman, H. R., and Hanks, E. N. 1981. Turnips and fodder radishes as forage crops for lambs. Wash. Agric. Exp. Stn. Bull. 09048.
15. Helson, G. A. H., and Norris, D. O. 1943. Transmission of potato virus diseases. 3. Susceptibility of Cruciferae to potato leafroll virus. J. Counc. Sci. Ind. Res. Aust. 16:261-262.
16. Kaniewski, W. K., and Thomas, P. E. 1988. A two-step ELISA for rapid, reliable detection of potato viruses. Am. Potato J. 65:561-571.
17. Pike, K. S., Allison, D., and Boydston, L. 1988. Wash. Aphid Bull. 19:1-3.
18. Powell, D. M., and Mondor, W. T. 1976. Area control of the green peach aphid on peach and the reduction of potato leafroll virus. Am. Potato J. 53:123-139.
19. Salaman, R. N., and Wortley, W. R. S. 1939. Potential hosts of potato viruses in garden and field. Nature 144:1049-1050.
20. Smith, H. G., and Hinckles, J. A. 1985. Studies on beet western yellows virus in oilseed rape (*Brassica napus* ssp. *oleifera*) and sugar beet (*Beta vulgaris*). Ann. Appl. Biol. 107:443-484.
21. Thomas, P. E. 1983. Sources and dissemination of potato viruses in the Columbia Basin of the northwestern United States. Plant Dis. 67:744-747.
22. Thomas, P. E., and Kaniewski, W. K. 1986. Overwintering of potato leafroll and beet western yellows viruses in winter annual weeds. (Abstr.) Phytopathology 76:847.
23. Thomas, P. E., Zielinska, L. M., and Smith, D. R. 1978. Virus diseases of potatoes in Washington, their costs and controls. Annu. Proc. Wash. Potato Conf. 17:59-64.
24. Washington Agricultural Statistics. 1986–1987. Washington Agricultural Statistics Service, Olympia. 108 pp.
25. Wright, N. S. 1970. Combined effects of potato viruses X and S on yield of Netted Gem and White Rose potatoes. Am. Potato J. 47:475-478.