Red Kidney Bean, a Useful Bioassay Host for Qualitative and Quantitative Work with Potato Virus M

C. Hiruki

Department of Plant Science, University of Alberta, Edmonton, Alberta, Canada.

Supported in part by Alberta Agricultural Research Trust Grant AR-55-28048. The author thanks R. Stace-Smith and R. H. Bagnall for supplying PVM, PVS, and PVM-antiserum.

Potato virus M (PVM) occurs naturally in several potato cultivars in America (2, 3, 9), Europe (3, 4, 10, 12), and Asia (7). The King Edward potato (*Solanum tuberosum* L.), which originated in England, is believed to be universally infected with the paracrinkle strain of the virus (10).

Nicotiana debneyi Domin. and *Datura metel* L. are useful in determining infectivity of PVM in extracts (2, 3). However, no detailed work has been published regarding the use of these or other species as quantitative assay hosts.

Bean (*Phaseolus vulgaris* L.), an efficient local lesion host for many plant viruses, has been neglected in the assay of PVM, probably because of varietal differences in its susceptibility to PVM. As the result of an extensive search for more suitable assay plants, Red Kidney bean was found in the present study to be the most susceptible among the 12 cultivars of bean tested under comparable conditions, and is useful for assaying PVM.

Virus extracts were prepared in 1% K_2HPO_4, pH 8.5, from leaves of young tomato (*Lycopersicon esculentum* Mill.) or of vigorously growing potato infected with the Alberta isolate (AP-1) of PVM. Well-expanded primary leaves of Red Kidney bean, grown at 17 ± 2 C, were dusted with 600-mesh Carborundum and inoculated by rubbing each half-leaf with a Q-tip dipped in inoculum.

The average numbers of local lesions from 12 half-leaves of 10 bean cultivars were: Red Kidney, 43.5; Bountiful, 32.4; Market Gardener's Wax, 32.4; Improved Golden Wax Dwarf, 28.9; Green Stringless Dwarf, 18.6; Top Notch Wax Dwarf, 15.2; Unrivalled Wax, 10.0; Kentucky Wonder Wax, 2.3; Kentucky Wonder Green, 2.3; and Stringless Black Valentine, 1.2. Somewhat diffused, brown local lesions developed on the primary leaves of these cultivars 5 to 6 days after inoculation. Blue Lake Stringless and Pinto were resistant to PVM.

Certain virus strains differ in their ability to infect bean or in the size of lesions produced (6, 8). To investigate this possibility with PVM, three isolates, AP-1 and one isolate each from British Columbia and New Brunswick (1, 2), were tested on Red Kidney bean using the half-leaf method. The results indicated no appreciable difference in time, nor in the size and appearance of local lesions (Fig. 1).

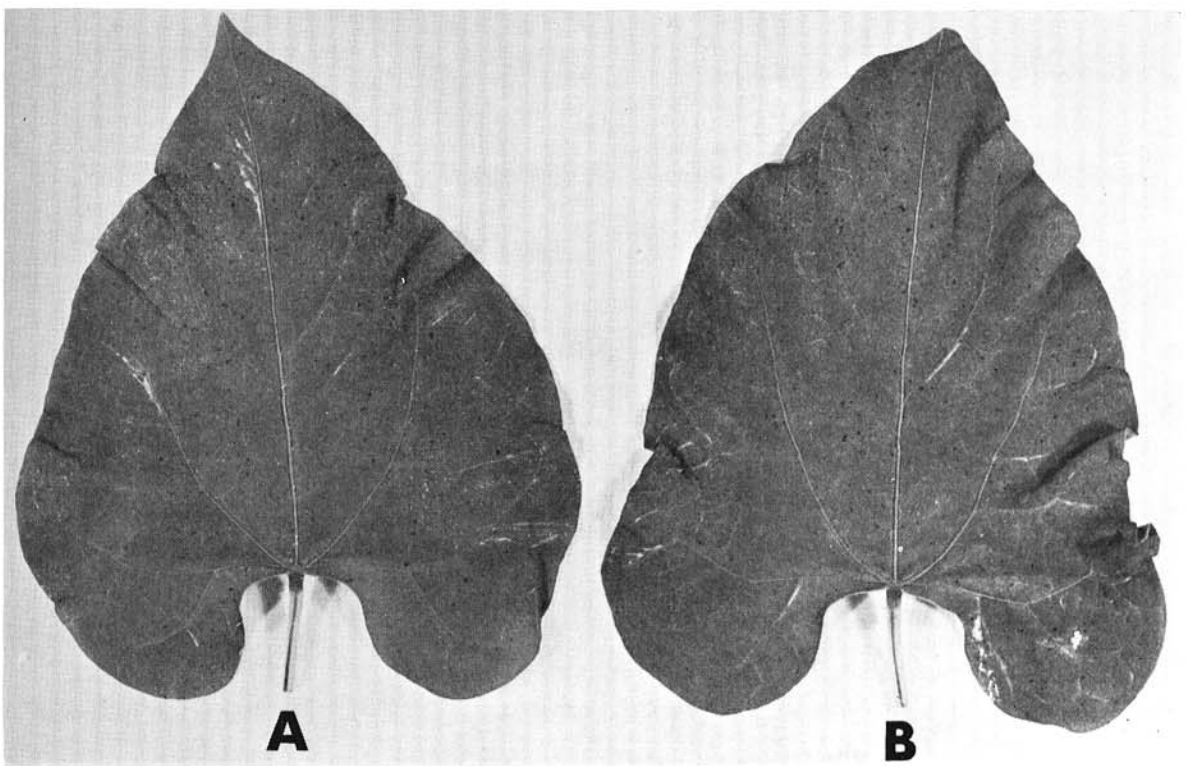

Fig. 1. A comparison of three isolates of potato virus M on leaves of *Phaseolus vulgaris* L. 'Red Kidney' 10 days after inoculation. **A)** Left half, an Alberta isolate (AP-1); right half, a British Columbia isolate. **B)** Left half, the AP-1; right half, a New Brunswick isolate.

TABLE 1. Effect of potato virus S (PVS), potato virus X (PVX), sap of *Nicotiana debneyi,* and sap of *Solanum tuberosum* on the development of local lesions in Red Kidney bean inoculated with potato virus M (PVM)

Inoculum	Experiment				
	A	B	C	D	E
	Ratio[a]				
PVM + 1% K_2HPO_4	100.0	100.0	100.0	100.0	100.0
PVM + PVS	3.5	15.4	10.8	1.0	7.3
PVM + PVX	29.0	81.3	78.3	53.6	31.7
PVM + *N. debneyi* sap	152.9	133.0	227.7	68.0	174.3
PVM + *S. tuberosum* sap	53.0	74.6	82.7	28.3	28.1

[a] Each figure represents the ratio of an average number of local lesions obtained on 12 half-leaves inoculated with mixtures of either PVM + PVS, PVM + PVX, PVM + *N. debneyi* sap, or PVM + *S. tuberosum* sap to number of lesions resulting from inoculation of PVM alone.

In distinguishing PVM and potato virus S (PVS), 7 plant species were used as differentials, of which *N. debneyi,* cowpea (*Vigna sinensis* Endl.), and guar (*Cyamopsis tetragonoloba* L.) were locally infected with PVM (1, 2). Lesions incited by PVS in guar were similar to those of PVM, but PVS had a longer incubation period (6-12 days) in comparison with PVM (4-6 days) (2, 3). Potato virus F also incited similar lesions in guar cotyledons (1, 2). Therefore, guar would not be considered a good differential host for PVM. Local lesions of PVM developed in Red Kidney bean were easily distinguishable in appearance and by a longer incubation period from those of alfalfa mosaic virus and of tobacco rattle virus, both of which were reported to occur in potato (5, 13).

Since most commercial potato varieties were infected with potato virus X (PVX) and PVS (11), the interference between PVM, PVX, and PVS was investigated in Red Kidney bean by using mixed inocula. PVM and PVX were increased in potato, and PVS in *N. debneyi.* Healthy leaves of these species served as controls. Two g each of infected and healthy leaves were separately ground in 2 ml 1% K_2HPO_4 and centrifuged at 3,000 *g* for 20 min. Each supernatant sample was diluted 1:5 in 1% K_2HPO_4, then mixed in a 1:1 ratio (Table 1). PVM infection was strongly interfered with by PVS in Red Kidney bean leaves, although these are generally accepted as separate viruses rather than strains (3). Upon mixing, sap containing PVX from infected potato leaves was inhibitory to PVM, but the extent of inhibition hardly exceeded that of sap from healthy potato leaves under the same conditions (Table 1), suggesting that the inhibition was not caused by PVX. This conclusion was supported by the results obtained in a separate experiment using purified PVX (Hiruki, *unpublished data*). Sap from healthy *N. debneyi* increased, in some unexplained manner, the infectivity of PVM in four of five tests. This apparent stimulatory effect appeared to vary according to the physiological conditions and age of *N. debneyi* leaves. These results suggest that the number of local lesions of PVM could be affected if PVM would be directly assayed in Red Kidney bean with extracts from existing commercial varieties of potato infected with PVS.

Since Red Kidney bean was immune to PVX and PVS, it was possible to sort out PVM from samples infected with these viruses and then subculture in tomato. Thus, Red Kidney bean is useful not only as a bioassay plant for quantitative work but also as a differential host for diagnostic purpose.

LITERATURE CITED

1. BAGNALL, R. H. 1960. Potato virus F latent in an imported variety and resistance to the virus in an interspecific potato hybrid. Phytopathology 50:460-464.
2. BAGNALL, R. H., R. H. LARSON, & J. C. WALKER. 1956. Potato viruses M, S, and X in relation to interveinal mosaic of the Irish Cobbler variety. Wis. Agr. Exp. Sta. Res. Bull. 198. 45 p.
3. BAGNALL, R. H., C. WETTER, & R. H. LARSON. 1959. Differential host and serological relationships of potato virus M, potato virus S, and carnation latent virus. Phytopathology 49:435-442.
4. BAWDEN, F. C., B. KASSANIS, & H. L. NIXON. 1950. The mechanical transmission and some properties of potato paracrinkle virus. J. Gen. Microbiol. 4:210-219.
5. CERVANTES, J., & R. H. LARSON. 1961. Alfalfa mosaic virus in relation to tuber necrosis in the potato variety Red La Soda. Wis. Agr. Exp. Sta. Res. Bull. 229. p. 37.
6. HIRUKI, C. 1965. Studies on strain relationships and variation in tobacco mosaic virus in Japan. I. The strains of tobacco mosaic virus isolated from field grown tomato plants. Hatano Tobacco Exp. Sta. Bull. 56:1-18.
7. HORIO, H., I. YANO, & K. ESUMI. 1969. Potato virus M found in Japan. Ann. Phytopathol. Soc. Japan 35:47-54.
8. HULL, R. 1968. Virus diseases of garden lupin in Great Britain. Ann. App. Biol. 61:373-380.
9. KAHN, R. P., H. A. SCOTT, J. BOZICEVICH, & M. M. VINCENT. 1967. Detection of potato viruses X, M, and S in dormant potato tubers by the bentonite flocculation test. Phytopathology 57:61-65.
10. KASSANIS, B. 1960. Potato virus M and paracrinkle. Nature 188:688.
11. STACE-SMITH, R., & F. C. MELLOR. 1968. Eradication of potato viruses X and S by thermotherapy and axillary bud culture. Phytopathology 58:199-203.
12. VULIĆ, M., & W. HUNNIUS. 1968. Die Reaktionen verschiedener Pflanzenarten auf Blattinfektionen mit S- und M-virus der Kartoffel. Phytopathol. Z. 59: 225-248.
13. WALKINSHAW, C. H., & R. H. LARSON. 1959. Corky ringspot of potato, a soil-borne virus disease. Wis. Agr. Exp. Sta. Res. Bull. 217. p. 31.