Effect of Potato Virus Y and Tobacco Mosaic Virus on Field-Grown Burley Tobacco

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ABSTRACT

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Yield, grade index, crop index, value per kilogram, and value per hectare were reduced when Burley 37 became infected with tobacco mosaic virus (TMV), potato virus Y (PVY), or both. A greater reduction was caused by PVY than by TMV, but the two viruses together caused the greatest reduction. Similarly, TMV+PVY caused greater variation from normal of chemical constituents than did either PVY or TMV alone. Compared to healthy tobacco, PVY-infected tobacco had less nicotine and TMV-infected tobacco had less α-amino nitrogen, but tobacco infected with TMV+PVY had less water-soluble acids, nicotine, phenols, and α-amino nitrogen and a lower percentage of its total nitrogen was soluble. Tobacco infected with TMV+PVY had more nitrate and insoluble nitrogen. Flowering was delayed in plants infected with TMV or TMV+PVY, but PVY infection alone had little effect on flowering.

Additional key words: *Nicotiana tabacum*, double infection, virus interactions, veinbanding.

Tobacco mosaic virus (TMV) and potato virus Y (PVY) are known to infect many species of plants, including several commercial crops. Losses of tobacco from these viruses have been estimated, but little is known about their effect on growth, yield, and quality of tobacco. Thomson and Wright (9) reported reductions in weight, leaf area, and smoking quality of flue-cured tobacco infected with PVY. A study in the greenhouse showed that plant height, plant weight, and leaf weight of susceptible cultivars were reduced by TMV and PVY, and that together they generally had a synergistic effect on a cultivar susceptible to both viruses (7). Other recent studies have determined the effect of TMV on the chemical characteristics of flue-cured tobacco (5) and the effect of early harvest on tobacco infected with PVY (8). The purpose of this study was to ascertain some of the effects of these viruses on production and on selected chemical constituents, primarily the nitrogenous components, total phenols, and aklakoids of burley tobacco.

MATERIALS AND METHODS

Burley tobacco (*Nicotiana tabacum* L. 'Burley 37') (Bu 37) was used because it is susceptible to both TMV and PVY and resistant to several other tobacco diseases. The test plots were four adjacent 42-plant rows in a Cumberland silt loam soil. Only plants in the center two rows of each plot were inoculated; the outer two rows served as barriers to minimize spread of the viruses to adjacent plots. The treatments consisted of inoculating plants with TMV, PVY, or TMV+PVY, or leaving them noninoculated. The treatments were replicated three times in a randomized block design. The end plants of each treatment row were not harvested; thus, data were obtained from 80 plants in each plot.

The experiment was conducted first in 1968 and was repeated in 1969. Seedlings were transplanted 28 May 1968 and 30 May 1969. The plants were inoculated 28 June 1968 and 26 June 1969, by rubbing two leaves per plant with a virus suspension prepared from systemically infected plants grown in the greenhouse. This inoculum was prepared by homogenizing one part infected plant material in a blender with about five parts 0.1% Na_2SO_3 (w/v), filtering the homogenate through cheesecloth, and adding silicon carbide, average particle size 43 μm (320-grit).

Normal cultural and handling practices were followed for growing, harvesting, and curing the tobacco. Flowering plants were counted on 14 August 1969.

The yield and grade index were determined from the cured tobacco. The crop index, value per kilogram, and value per hectare were calculated for each treatment. The grade and crop indices were obtained as previously described (6). Composite samples of the cured leaf were analyzed for selected constituents utilizing the following methods: α-amino nitrogen, pH, total nitrogen, and

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water-soluble acids were determined by, respectively, methods 2, 20, 33, and 38 as described by Bacot (2). Determination of nitrate nitrogen was by the method of Collins et al. (3), that of total phenols by the method of Andersen and Todd (1), and that of nicotine by Griffith's method (4). Acid-insoluble nitrogen was determined by measuring the total nitrogen after extraction with 0.5% acetic acid (Mitchem, *personal communication*). The percent of the total nitrogen that was soluble was calculated by the formula:

$$\frac{\text{(Total nitrogen)} - \text{(acid-insoluble nitrogen)}}{\text{(Total nitrogen)}} \times 100$$

(Apparent discrepancies in the percent total nitrogen that was soluble are due to rounding). The data were analyzed statistically by an analysis of variance.

RESULTS

Plants inoculated with TMV+PVY became chlorotic and were stunted. During the growing season those plants (approximately 5%) which developed symptoms not consistent with the treatment were removed and discarded. The data on flowering, yield, and quality are given in Table 1. The chemical data are given in Table 2.

Flowering.—When the count was made, more noninoculated than inoculated plants were flowering. No significant reduction in percentage of flowering was attributable to PVY, but TMV, either alone or with PVY, caused a highly significant reduction.

Yield.—Both TMV and PVY significantly reduced yield, but the reduction was greater with PVY (about 35%) than with TMV (about 28%). In combination, the two viruses caused an additive reduction in yield: about 64%.

Grade index.—Both viruses significantly reduced the grade index, but PVY caused a greater reduction than TMV. The tobacco from plants infected with both viruses had the lowest grade index, but it was not significantly lower than that of tobacco from plants infected with PVY alone.

Crop index.—The crop index was significantly reduced by each virus and by the two viruses combined. Reductions by TMV, PVY, and TMV+PVY were about 47, 62, and 82%, respectively.

Value per kilogram.—Plants infected with the two viruses, either individually or combined, produced leaves of considerably lower value than did healthy plants. Again, the reduction was greatest for the two viruses together, but in this case it was more than additive: TMV reduced the value per kg by $.035, PVY caused a reduction of $.085, and the two viruses together reduced the value by $.148.

Value per hectare.—Each virus singly and the two together significantly reduced the value per hectare. The value was reduced more by PVY than by TMV, and the reduction by the combined viruses was approximately equal to the sum of the reductions caused by the individual viruses.

Chemical constituents.—Compared to noninoculated plants, the composition of PVY-infected plants differed

TABLE 1. Effect of potato virus Y (PVY) and tobacco mosaic virus (TMV) on yield and quality of Burley 37 tobacco[z]

Treatment	Flowering (%)	Yield (kg/ha)	Grade index	Crop index	Value	
					$/kg	$/ha
TMV	16.4 b	1817 b	0.399 b	566 b	1.558 b	2,831 b
TMV+PVY	15.0 b	909 c	0.253 c	194 d	1.445 d	1,314 d
PVY	24.8 a	1626 b	0.309 c	406 c	1.508 c	2,453 c
None	29.0 a	2520 a	0.539 a	1066 a	1.593 a	4,014 a

[z]All data except flowering are 2-yr means of three replications per year. The values for percentage flowering are means for one count of three replications made on 14 August 1969. In each column, means followed by the same letter are not significantly different ($P = 0.05$) according to Duncan's multiple range test.

TABLE 2. Effect of tobacco mosaic virus (TMV) and potato virus Y (PVY) on selected chemical characteristics of Burley 37 tobacco[y]

Virus treatment	Nitrogen					Total phenols (%)	Nicotine (%)	Water-soluble acids (ml)[b]	pH
	Total (%)	Acid-insoluble (%)	Percent of total soluble (%)	α-Amino (%)	Nitrate (%)				
TMV	4.43 a	1.71 a	61.3 a	0.596 b	0.22 a	0.78 ab	4.76 ab	4.30 a	5.3 a
TMV+PVY	4.51 a	2.43 b	45.6 b	0.488 c	0.33 b	0.60 b	3.66 c	3.34 b	5.5 a
PVY	4.42 a	1.72 a	61.0 a	0.681 a	0.25 ab	0.76 ab	4.31 b	3.96 ab	5.4 a
None	4.40 a	1.34 a	69.4 a	0.645 a	0.17 a	1.08 a	4.96 a	4.57 a	5.3 a

[y]Each number represents the mean of three replications. In each column, means followed by the same letter are not significantly different ($P = 0.05$) according to Duncan's multiple range test.

[z]Milliliters of 0.1 N NaOH required to neutralize 1.0 g tobacco.

significantly only in that nicotine was reduced by about 13%. The only constituent significantly affected by TMV was α-amino nitrogen, which was reduced about 8%. However, TMV+PVY significantly reduced the percentages of water-soluble acids, nicotine, phenols, α-amino nitrogen, and the total nitrogen that was acid-soluble, and significantly increased the percentages of nitrate and acid-insoluble nitrogen.

DISCUSSION

The yield, value, and grade and crop indices of Bu 37 were reduced more by PVY than by TMV, and still more by the two viruses combined. These findings confirm those of the previous greenhouse study (7) in which PVY generally affected Bu 37 more than TMV did and the two viruses acted synergistically.

Either alone or with TMV, PVY had no significant effect on flowering, but TMV alone significantly delayed flowering. This finding was unexpected because plants infected with PVY appear to mature earlier than healthy tobacco and, if left in the field, often turn yellow, then brown, and become desiccated before healthy tobacco is ready to be harvested. However, certain chemical constituents do not reach levels comparable to those in healthy tobacco until the normal harvest dates (8). Thus, as an indicator of maturity, the flowering status of PVY-infected plants seems to be better than the appearance of their leaves.

All of the chemical characteristics that were measured, except total nitrogen and pH,were affected by one or both viruses, although the changes were not always significant. Nitrate and acid-insoluble nitrogen generally increased in the plants infected with either or both viruses, whereas, the other constituents generally decreased. As might be expected, the effects of TMV+PVY were greater than the effects of either PVY or TMV alone.

Tobacco mosaic virus is considered a threat to tobacco production because of its persistence and the ease with which it is transmitted mechanically, but PVY actually poses a more serious threat. There are cultivars available with resistance to TMV, but none with resistance to PVY. Furthermore, PVY causes greater losses in yield and quality than does TMV. In this experiment PVY reduced the value per hectare (which reflects the reductions in both weight and quality) by about 40%, whereas TMV reduced the value per hectare by only 30%. In some burley cultivars the loss in value per hectare from PVY exceeds 75% (Sievert, *unpublished*). In their study of flue-cured tobacco, Thomson and Wright (9) found PVY reduced the weight of cured leaf by 28.5%. In this study PVY reduced the yield by 35.5%.

For several reasons the losses reported in this study may be greater than would be generally experienced in natural infections. First, inoculation produced uniform infection early in the growing season; under natural conditions, infection occurs sporadically throughout the growing season. Second, all the plants became infected, which might not occur naturally. Third, simultaneous infection by two viruses is unusual. Even the recovery of two viruses from a single diseased plant is uncommon, although there were several such instances in Virginia flue-cured tobacco in 1976 (J. Reilly, *personal communication*). Nevertheless, this study gives an indication of the losses that might occur should PVY become epiphytotic in burley tobacco.

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