Effect of Tobacco Etch and Tobacco Vein Mottling Virus on Yield of Burley Tobacco Genotypes

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ABSTRACT

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Burley tobacco (*Nicotiana tabacum* L.) cultivars and experimental breeding lines were evaluated for yield response in the presence of tobacco etch (TEV) and tobacco vein mottling (TVMV) viruses under field conditions for 2 years at two locations. Breeding lines Greeneville 131 (from Tennessee) and MDH 5, MDH 19, MDH 25, and MDH 28 (from North Carolina) were found to be highly resistant to TEV and TVMV. These breeding lines exhibited little or no virus symptoms and their resistance is conditioned by a single recessive factor derived from Virgin A Mutant. Cultivars Kentucky 14 and Kentucky 10 exhibited high yields despite presence of virus symptoms. High-yielding cultivars R7-11, Kentucky 14 × L8, and Burley 21 × Kentucky 10 had only moderate yields in the presence of either virus. Cultivars Burley 37 and Burley 49 were extremely susceptible, based on very low yields and severe virus symptoms. Cultivars Havana 307 and Sota 6505 were found to possess a new source of TEV and TVMV resistance. It may be possible to combine different sources of resistance along with tolerance in a single genotype.

Tobacco etch (TEV) and tobacco vein mottling (TVMV) viruses (both members of the potyvirus group) cause significant losses on burley tobacco (*Nicotiana tabacum* L.) over the entire burley tobacco-producing region of the United States which includes North Carolina, Virginia, Kentucky, and Tennessee (2,4–7,14,15). Incidence of TEV and TVMV vary greatly from year to year (2,3,5), presumably due to shifts in populations of their aphid vectors, but may be also influenced by other factors, such as amount of overwintering inoculum and environmental effects. Thus, the amount of damage caused by these viruses fluctuates from year to year. For example, incidence of TVMV and TEV in North Carolina in 1984 was approximately 27 and 7%, respectively, causing an estimated loss of $2 million (11). In 1986, incidence was 0.7 and 2% for TVMV and TEV, respectively, causing a combined loss of less than $1 million (10).

Currently, control of TEV and TVMV on tobacco is achieved primarily through the use of resistant or tolerant cultivars (5). A new cultivar, Tennessee 86, with high resistance to both TEV and TVMV, was released in 1986 (12). Before this release, there were no resistant cultivars in commercial production. However, significant differences for tolerance to TEV among burley tobacco cultivars were reported in 1970 (4).

The present study was initiated in 1984 to evaluate the relative tolerance or resistance of burley tobacco cultivars and breeding lines to TEV and TVMV under field conditions. In this paper, the term resistance is defined as any inherited characteristic of a host genotype that lessens the effects of either virus (i.e., resistant plants reduce the severity of virus symptoms and are less damaged than are susceptible plants) (18). In contrast, a tolerant genotype will be defined as one that exhibits virus symptoms, is attacked to the same degree as a susceptible genotype, but suffers less damage (in terms of yield and quality) as a result of virus infection (18). The main objectives of the experiment were to determine yield (kg/ha) and quality ($/ha) of germ plasm in the presence and absence of each virus separately and to estimate percent reduction in yield and dollar value due to virus infection.

MATERIALS AND METHODS

Evaluations for resistance or tolerance to TEV were performed during 1984 and 1985 at two locations: Tobacco Experiment Station, Greeneville, TN, and Mountain Research Station, Waynesville, NC. The 1985 Waynesville experiment was not harvested due to drought damage. Tolerance to TVMV was also evaluated in 1984 and 1985 at two locations: Mill Ridge, NC, and the Upper Mountain Research Station, Laurel Springs, NC. A severe freeze in early September killed the plants in the 1985 test at Laurel Springs and it was not harvested.

For each test, the experimental design consisted of a split-plot experiment with three replications. Inoculated vs. non-inoculated treatments were assigned to whole plots and genotypes (entries) to subplots. Bartlett's tests for homogeneity of variance were performed and no evidence of heterogeneous variances was detected. Therefore, data were pooled across years and locations. Because data from some locations were lost due to

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drought or early frost, we used the available year/location means (labeled as environments) in the combined analyses.

Genotypes (entries) were selected on the basis of their popularity and widespread use in commercial production (includes both inbred lines and F1 hybrids) or because of their potential as sources of TEV or TVMV resistance or tolerance. Commercial cultivars tested can be grouped into high-, moderate-, and low-yielding groups in the absence of virus infection (1,9). High-yielding U.S. commercial cultivars were Kentucky 14 (Ky 14), R7-11, Kentucky 14 × L8 (Ky 14 × L8), and Burley 21 × Kentucky 10 (Bu 21 × Ky 10). Moderate-yielding U.S. cultivars included Kentucky 10 (Ky 10), Kentucky 15 (Ky 15), Kentucky 17 (Ky 17), Virginia 509 (Va 509), Virginia 528 (Va 528), Coop 313, Clays 501, Burley 21 (Bu 21), and Burley 21 × L8 (Bu 21 × L8). Low-yielding cultivars include Burley 37 (Bu 37), Burley 49 (Bu 49), and Burley 37 × L8 (Bu 37 × L8). European cultivars (moderate yield) Jaraiz 1 and MB(J) from Spain and Sota 6505 from Switzerland were also included. We also evaluated two low-yielding lines used by tobacco breeders as sources of resistance to TEV, TVMV, and potato virus Y (PVY): Virgin A Mutant (VAM), which possesses a single recessive gene conditioning resistance to all three viruses (5,8), and Havana 307 (HA 307), a cigar-wrapper tobacco. The remaining entries are experimental breeding lines that possess a recessive factor for TEV and TVMV resistance derived from VAM. These lines were Greeneville 131 (GR 131) from the Tennessee breeding program, and MDH 5, MDH 17, MDH 19, MDH 25, MDH 28, and NC 107 from the North Carolina breeding program. MDH lines are maternal doubled haploids derived from the cross Ky 14 × NC 107.

Seed of all genotypes were sown in plant beds in March and seedlings were transplanted to the field between 25 May and 6 June of each year. Field plots consisted of single rows of 22–30 plants spaced 46 cm apart within the row and 122 cm between rows. Standard cultural practices for burley tobacco were used in every experiment.

Virus inoculations were performed 3–4 wk after transplanting using an artist's airbrush. A strain of TEV that is not aphid transmissible (isolate NC 191) was used in all TEV experiments. The isolate of TVMV was NC 148 (8). Inoculum preparation was as previously reported (4).

Competitive (i.e., bordered) plants within plots were harvested at maturity and air-cured using conventional practices. Cured-leaf yields (kg/ha) were determined and quality ($/ha) was assessed using certified U.S. government grades.

Table 1. Analyses of variance for yield and dollar value/ha of burley tobacco genotypes evaluated for reaction to tobacco etch or tobacco vein mottling virus

		Mean squares			
		Yield (kg/ha)		Value ($/ha)	
Source	df	TEV	TVMV	TEV	TVMV
Environments (E)	2	33,221,148.3 **[a]	6,267,452.8 **	103,280,629.2 **	24,941,725.2 **
Virus treatment (V) (inoculated vs. noninoculated)	1	567,129.7	443,642.5	19,069,572.5	1,489,342.6
E × V	2	578,882.7	125,534.4	1,947,439.7	468,650.3
Genotypes (G)	28	459,015.5 **	1,367,766.9 **	1,945,035.4 **	5,154,440.5 **
V × G	28	70,861.7	113,878.5	252,716.0	411,453.8
Error	112	115,719.4	91,885.7	475,595.5	551,279.3

[a] ** = Significant at the 0.01 probability level.

Table 2. Mean yield and dollar value of burley tobacco genotypes infected with tobacco etch virus[a]

Rank	Entry	Yield (kg/ha)	Value[b] ($/ha)
1	Greeneville 131	2,982.6	11,721.4
2	MDH 25	2,949.3	11,590.5
3	MDH 5	2,935.5	11,791.8
4	Kentucky 14	2,913.3	11,481.3
5	MDH 28	2,705.4	10,594.3
6	Kentucky 10	2,704.6	10,669.7
7	MDH 19	2,686.3	10,636.3
8	Kentucky 15	2,643.0	10,339.4
9	R7-11	2,334.3	10,317.7
10	Burley 21 × Kentucky 10	2,602.9	10,097.9
11	Burley 21	2,578.0	10,137.6
12	Kentucky 14 × L8	2,533.8	9,959.8
13	Coop 313	2,506.6	9,822.0
14	Sota 6505	2,443.6	9,607.6
15	Jaraiz 1	2,417.0	9,418.6
16	Virginia 509	2,411.4	9,452.7
17	MDH 17	2,383.0	9,314.9
18	Virginia 528	2,370.7	9,276.1
19	Kentucky 17	2,310.0	9,069.8
20	MB(J)	2,303.8	8,988.8
21	Burley 21 × L8	2,290.6	8,969.8
22	Clays 501	2,288.2	9,014.8
23	MDH 9	2,286.3	8,907.6
24	VAM	2,254.0	8,835.7
25	Burley 37 × L8	2,187.7	8,585.7
26	Havana 307	2,178.8	6,058.2
27	NC 107	2,085.1	8,215.2
28	Burley 37	1,858.1	7,221.5
29	Burley 49	1,671.0	6,460.8
	$LSD_{0.05}$	435.8	1,985.6

[a] Data represent means of three environments (Greeneville, TN, in 1984 and 1985, and Waynesville, NC, in 1984).
[b] Weighted average dollar value/kg × kg/ha.

RESULTS

The pooled analyses of variance for yield and dollar value per hectare for TEV and TVMV experiments indicated highly significant differences among environments ($P = 0.01$) (Table 1). No significant differences ($P = 0.05$) due to inoculation (virus treatments) were detected with either virus due to contamination of noninoculated plots. Yield and value ($/ha) reduction due to virus inoculation as percentage of healthy controls could not be computed from our data because of the absence of significant differences between inoculated and noninoculated plots in the combined analyses (Table 1). Differences among genotypes for reaction to TEV or TVMV were highly significant ($P = 0.01$). Interaction terms (virus treatment × genotypes) were not statistically significant ($P = 0.05$) (Table 1).

Yield and quality measurements of cultivars and breeding lines infected with TEV and TVMV are given in Tables 2 and 3, respectively. Breeding line GR 131 was the highest yielding entry and had the largest dollar value in the presence of either virus. Cultivars Bu 37 and Bu 49 were the most susceptible to both viruses based on extremely low yields (significantly lower than in the absence of virus) (1,9) and severe virus symptoms. Several MDH lines performed well when inoculated with either virus (e.g., MDH 5, MDH 19, MDH 25, and MDH 28) (Tables 2 and 3).

Cultivars Ky 10 and Ky 14 exhibited

the highest degree of tolerance to both viruses. These cultivars lack the single gene for TEV and TVMV resistance from VAM present in experimental breeding lines (Tables 2 and 3), but still yield relatively well in the presence of either virus. In contrast, cultivars R7-11, Ky 14 × L8, and Bu 21 × Ky 10, which yield very well in the absence of virus infection (1,9), are intolerant or susceptible and show yield repression when infected with either virus. Extremely susceptible cultivars Bu 37 and Bu 49 exhibited severe yield reductions relative to their performance in the absence of either virus (1,9). Both of these cultivars were more sensitive to TVMV than TEV.

Two entries showed promise as new sources of resistance to both TEV and TVMV: Sota 6505 and HA 307. Resistance of HA 307 to PVY, TEV, and TVMV (under greenhouse conditions) had been reported previously (8). Yield of HA 307 and Sota 6505 in this experiment was similar to that observed in other trials in the absence of either virus (authors, *unpublished*). HA 307 showed a low quality value because it is a cigar-wrapper tobacco and quality was measured using burley tobacco quality standards. Plants of both Sota 6505 and HA 307 were almost entirely devoid of virus symptoms.

DISCUSSION

The magnitude of environmental effects in combined analyses of TEV and TVMV experiments was much greater than any other source of variation. These differences are not surprising and may be the result of temperature differences and other environmental factors known to affect virus disease severity (17). For example, TEV experiments were conducted at two locations with very different climatic conditions. The Greeneville, TN, location experiences much warmer temperatures than the Waynesville, NC, location because of the higher elevation of the latter. Differences in light quality and intensity, rainfall, soil types, and seasonal changes may also account for observed differences among environments.

The absence of significant differences between whole-plot treatments (inoculated vs. noninoculated plots) was not completely unexpected because no practical means were available to prevent infection of plants in noninoculated plots via aphids from indigenous sources of inoculum. Both TEV and TVMV overwinter in several weed hosts and are vectored by several different aphid species (7). Plot contamination by nontarget viruses was believed to be insignificant, however, because the test locations were chosen based on the natural occurrence and preponderance of the viruses under study (5). We had hoped that uniform infection of the inoculated plots early in the season when compared with later natural infection of noninoculated plots would provide a quantitative estimate of the damage caused by viral infection. Early infection by another potyvirus, PVY, had previously been demonstrated to adversely affect yield more than later infection (19), but apparently this was not true of TEV and TVMV. Definitive data on the effect of time of inoculation with TEV or TVMV on plant yield would be valuable information.

Because of the above problems, it was not possible to compute percent yield and quality reduction of inoculated genotypes relative to noninoculated controls in this experiment. The data we obtained permitted us only to evaluate the relative performance of the different tobacco genotypes when infected with either TEV or TVMV. Among commercial cultivars without specific genes for resistance, we observed apparent cases of tolerance (e.g., Ky 14 and Ky 10). These cultivars showed distinct virus symptoms such as mottling, vein banding, and necrotic flecks, yet their yield seemed to be relatively unaffected. Relative to the performance of cultivars in the absence of virus infection (1), there was some reduction in yield (less than 10%) and quality in apparently tolerant cultivars, but not nearly to the degree exhibited by highly susceptible cultivars such as Bu 37 and Bu 49. In the presence of TEV- or TVMV-susceptible cultivars, Bu 37 and Bu 49 showed very low yields, severe leaf and stem necrosis, and severe stunting. In some tests, there was little or no harvestable product at the end of the season.

In the presence of either TEV or TVMV, most experimental breeding lines possessing the single, recessive factor for virus resistance from VAM outperformed commercial cultivars with no specific genes for resistance (Tables 2 and 3). The VAM gene clearly protects against severe damage by these viruses.

Although breeding line GR 131 was consistently the most resistant genotype to both viruses, not all genotypes resistant or tolerant to one virus are necessarily resistant or tolerant to the other. Breeding line MDH 9 was far more sensitive to TEV (rank 23; Table 2) than to TVMV (rank 5; Table 3), whereas the relative rankings were reversed for cultivar Bu 21 × Ky 10. Thus, deployment of a resistant or tolerant cultivar requires knowledge of the identity of the virus causing the problem at a given location.

The resistance exhibited by the cultivars HA 307 and Sota 6505 will be investigated further. These cultivars offer a promising alternative to the VAM source of resistance to TEV and TVMV. New sources of resistance to TEV and TVMV are being sought because the

Table 3. Mean yield and dollar value of burley tobacco genotypes infected with tobacco vein mottling virus[a]

Rank	Entry	Yield (kg/ha)	Value[b] ($/ha)
1	Greeneville 131	3,084.3	11,856.0
2	MDH 19	2,939.4	11,451.6
3	MDH 5	2,915.9	11,003.9
4	Kentucky 10	2,866.4	11,097.2
5	MDH 9	2,804.3	10,785.3
6	Kentucky 14	2,781.0	10,651.4
7	MDH 25	2,731.3	10,426.0
8	Sota 6505	2,726.6	10,444.9
9	MDH 28	2,721.3	10,573.3
10	MDH 17	2,620.0	10,017.1
11	R7-11	2,500.7	7,516.7
12	Kentucky 14 × L8	2,500.4	9,623.1
13	Coop 313	2,465.5	9,618.9
14	Virginia 528	2,418.1	9,452.2
15	NC 107	2,373.3	9,223.7
16	Kentucky 15	2,306.1	8,681.6
17	Jaraiz 1	2,299.6	8,903.6
18	Burley 21 × Kentucky 10	2,296.9	8,838.4
19	MB(J)	2,242.8	8,662.8
20	Burley 21	2,089.0	8,147.8
21	VAM	2,020.8	7,706.4
22	Clays 501	1,979.9	7,578.7
23	Havana 307	1,923.0	4,383.8
24	Burley 37 × L8	1,885.5	6,934.0
25	Kentucky 17	1,836.8	7,007.9
26	Burley 21 × L8	1,823.9	6,804.9
27	Virginia 509	1,815.5	6,809.3
28	Burley 37	920.6	3,266.1
29	Burley 49	919.9	3,133.7
	$LSD_{0.05}$	388.4	2,097.9

[a] Data represent means of three environments (Mill Ridge, NC, in 1984 and 1985, and Laurel Springs, NC, in 1984).
[b] Weighted average dollar value/kg × kg/ha.

VAM factor is associated with severe susceptibility to chewing insects (20) and extreme susceptibility to tobacco blue mold (*Peronospora tabacina* Adam) (16), which is a potentially serious fungal disease of tobacco. Cultivar Tennessee 86 and breeding line GR 131 have been found to be relatively insensitive to insect damage even though they possess the VAM factor (12,13). It is possible that the linkage between virus resistance and insect susceptibility has been broken in these lines. All MDH lines appeared to be sensitive to chewing insects (*unpublished*) and this limits their potential as cultivars. No exceptions have been found to the association between blue mold susceptibility and the virus resistance factor from VAM. We have tested HA 307 for its reaction to tobacco blue mold, and it does not appear to be more susceptible than other cultivars (8). We have not tested Sota 6505 for blue mold sensitivity, but this cultivar has been grown commercially in Europe without reports of extreme susceptibility.

In summary, new sources of resistance to TEV and TVMV have been identified and tolerance to these viruses in commercial cultivars has been determined. It may be possible to combine tolerance with resistance in a single genotype which could provide enhanced protection of burley tobacco against TEV and TVMV.

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