

Soybean Mosaic Virus: Effects of Primary Disease Incidence on Yield and Seed Quality

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ABSTRACT

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Various percentages of seedling plants of the soybean cultivar Amsoy 71 were inoculated in a random pattern with soybean mosaic virus (SMV). Seeds harvested from plots, graded for seed-coat mottling, and analyzed for SMV antigen by solid-phase radioimmunoassay showed that levels of virus antigen were positively correlated with disease incidence. Yield reductions were similar in plots where 30 or 50% of the plants were inoculated. Experiments in which seed with known antigen levels was used to plant plots showed that antigen levels from seeds harvested from such plots either decreased or increased, depending on the year. It is probable that such results are dependent on the degree of secondary spread and time of inoculation.

Primary inoculum of soybean mosaic virus (SMV) in Iowa is infected soybean seedlings derived from infected seed (8). Secondary spread occurs by activity of several aphid species that transmit the virus in a nonpersistent manner (9). Reports documenting yield loss caused by SMV have been based on results from plots in which 100% of the plants were mechanically inoculated (3,10-12) with virus or plots in which aphid spread mediated inoculation of closely related resistant and susceptible lines (13) or blends of resistant and susceptible lines (14).

The use of infected seed results in random distribution of primary inoculum, with secondary spread resulting in infection of plants at diverse times (8). The objective of this research was to measure the effects that different levels of randomly distributed primary inoculum have on soybean yield and seed quality.

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MATERIALS AND METHODS

Isolate Ia 75-16-1 of SMV used in these experiments has been described previously (5,6,9). Experiments were conducted at two locations near Ames, IA. One field (location A) was bordered by woods to the south and grassy pasture on the other sides; the other field (location B) was surrounded by large soybean acreages.

All plots, arranged in a completely randomized design, were planted with certified seed of the soybean cultivar Amsoy 71 shown to be free of SMV by SPRIA (2). Each plot consisted of 300 plants arranged in three rows 6.1 m long and 76 cm apart. Plots were surrounded by a 9.2-m border sown with Amsoy 71. Planting dates were 16 May, 8 May, 7 June, and 27 May for 1980, 1981, 1982, and 1983, respectively.

In 1980, 0, 5, or 10% of the seedling plants in each of six plots at each location were randomly selected by computer and mechanically inoculated with SMV at growth stage V-1 (4). In 1981, 1982, and 1983, 0, 30, or 50% of the plants in each of six plots at two locations were randomly selected by computer and inoculated, except in 1983, when five plots each at location A were used. To estimate the final level of infection in each plot, leaves from 10% of the plants in each plot were selected by computer for random sampling and indexed for SMV antigen by enzyme-linked immunosorbent assay (ELISA) (7) at growth stage R-6. Seeds from each plot were harvested, dried to uniform moisture, and weighed. Two 100-seed samples from each plot were graded for seed-coat mottling and assayed by solid-phase radioimmunoassay (SPRIA) to detect virus antigen (2). Levels of SMV antigen were determined by linear interpolation from a standard curve, established concurrently with each

assay, which related SMV antigen to counts per minute. Data on yield, percentage of seeds with mottled seed coats, and nanograms of virus antigen

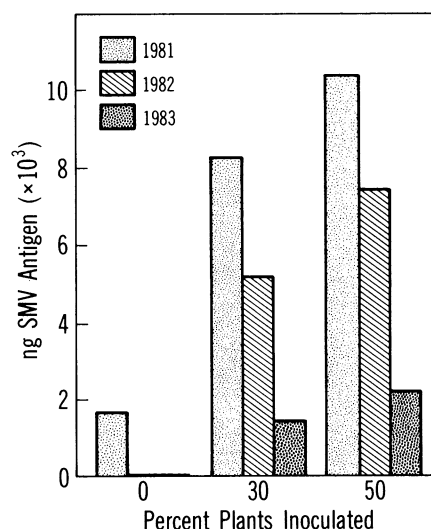


Fig. 1. Levels of soybean mosaic virus antigen detected by solid-phase radioimmunoassay in seeds harvested from plots where 0, 30, or 50% of the plants were randomly inoculated at two locations in 1981-1983. Data, which are the mean values from six replicates at each of two locations, except for 1983, when five replicates were used at one location, were combined because no significant location effect was detected by analysis of variance.

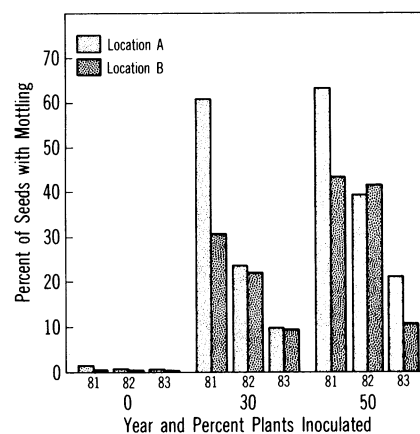


Fig. 2. Percentage of seeds with mottled seed coats harvested from plots where 0, 30, or 50% of the plants were randomly inoculated with soybean mosaic virus at two locations (A and B) in 1981-1983. Data are the mean values of six replicates, except for 1983, when five replicates were used at location A.

Table 1. Analysis of variance for the effects of year, location, and percentage of inoculated plants on seed-coat mottling, nanograms of virus antigen, seed weight, and deviation from expected number of infected plants in the absence of virus spread under three levels of inoculation with soybean mosaic virus

Source of variation	df	Mean squares			
		Percentage of seed-coat mottling	Nanograms of virus antigen	Seed yield (wt)	Deviation from expected no. of infected plants
Year (Y)	2	6,148.5*** ^a	267,509,556.0***	3,805,353.5***	200.3***
Location (L)	1	1,749.2***	30,604,917.7	14,916,274.7***	37.5
Y × L	2	582.1***	5,353,666.4	3,865,650.4***	86.9*
Error (a)	29	39.3	9,728,808.5	293,918.8	19.0
Percent plants inoculated (I)	2	12,970.3***	369,358,878.5***	943,085.2**	105.2***
I × Y	4	1,433.4***	38,004,314.7*	138,495.2	66.7**
I × L	2	321.2***	10,438,964.6	255,847.4	52.3
I × Y × L	4	236.1***	5,204,997.3	188,028.4	19.2
Error (b)	58	29.3	13,000,441.5	141,782.7	16.7

^aSignificant at * = $P = 0.05$, ** = $P = 0.01$, and *** = $P = 0.001$.

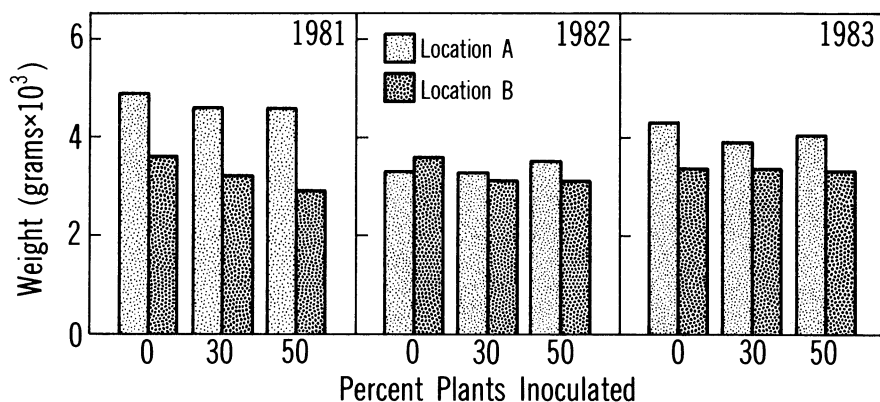


Fig. 3. Yield of soybean seed harvested from plots where 0, 30, or 50% of the plants were randomly inoculated with soybean mosaic virus at two locations in 1981–1983. Data are the mean values of six replicates, except for 1983, when five replicates were used at location A.

Table 2. Deviation from expected number of plants (assuming no virus spread) infected with soybean mosaic virus in plots where 0, 30, or 50% of the plants were randomly inoculated in 1981–1983^a

Percentage of plants inoculated	Year		
	1981	1982	1983
0	0.33	0.58	0.72
30	6.25	2.83	0.27
50	4.83	-1.67	-3.64

^aThe expected number of plants infected in plots where 0, 30, or 50% of 300 plants are inoculated is 0, 10, or 15, respectively, assuming no virus spread.

were analyzed with standard analysis of variance procedures. The chi-square test of heterogeneity was used to assess uniformity of response to the treatments applied to the plots across the experimental field.

In a related experiment, in successive years, we studied the effects of planting soybean seed containing known levels of virus antigen on final levels of virus antigen. Five seed lots with known levels of SMV antigen, as determined by SPRIA (2), were obtained by inoculating the soybean cultivar Amsoy 71 with SMV in 1979. Each seed lot was planted in four replicated plots of four rows 6.1 m long and 76 cm apart. Seed lots with known

SMV antigen levels from the 1980 harvest were used to plant plots in 1981, and similarly, seed of the 1981 harvest was used to plant the 1982 plots. Six and five replicates were planted with each seed lot in 1981 and 1982, respectively. Plots were arranged in a completely randomized design in all years. Planting dates were 29 April, 21 May, and 11 May in 1980, 1981, and 1982, respectively. The center two rows of each plot were harvested and dried to uniform moisture, and two 100-seed samples from each plot were assayed for SMV antigen by SPRIA (2).

RESULTS

Preliminary experiments in 1980 in

which 0, 5, or 10% of the plants in a plot were inoculated with SMV suggested that field variability was too great to obtain measurable differences in yield. Therefore, in 1981–1983, inoculum levels were increased to 0, 30, and 50%.

Levels of SMV antigen (nanograms) in seeds harvested from plots at the two locations were not significantly different (Table 1). Highest levels were detected in 1981, with decreasing amounts in 1982 and 1983. Antigen content was positively correlated ($r = 0.98, 0.99, \text{ and } 0.99$ for 1981, 1982, and 1983, respectively) with inoculum level (Fig. 1). Detection of virus antigen in the 0% inoculated plot in 1981 suggested that SMV spread into that plot from exterior sources in that year. This is consistent with other data presented in this study, suggesting that the greatest amount of secondary spread of SMV occurred in 1981.

The percentage of seed-coat mottling followed a pattern similar to the level of virus antigen in all three years (Fig. 2), although the amount of mottling was significantly greater at location A than at location B (Table 1). This apparent relationship is consistent with previous data showing that seed produced by SMV-infected Amsoy 71 has the unusual property that the percentage of seed-coat mottling is positively correlated with SMV antigen content (3).

Soybean yields differed significantly among years (Table 1); highest yields occurred in 1981, followed by 1983, then 1982. At each location, reductions in yield were generally similar in plots in which 30 or 50% of the plants were randomly inoculated, except for location A in 1982 (Fig. 3).

Because of the way the experiment was designed and executed, the expected number of plants infected at harvest can be calculated (Table 2). Additionally, because each level of inoculation was replicated six times in each year and location, we were able to test for lack of homogeneity within the experimental area. Because plants for inoculation at a

given level (i.e., 0, 30, or 50%) within the plot were randomly chosen and 10% of the 300 plants within each plot were randomly selected for ELISA at stage R-6, the expected frequency of a sampled plant being infected is $P(I) \times P(S)$, where $P(I)$ is the probability that a plant was inoculated and $P(S)$ is the probability that a plant was indexed for SMV infection by ELISA. The chi-square test of heterogeneity was therefore used to compare the agreement of the expected with the observed number of infected plants as determined by ELISA. The heterogeneity test suggested that, with the exception of plants inoculated at the 30% level in 1982 at one location, all plots treated in an identical manner in any given year and location responded in a similar manner (Table 3). The reason for the single aberration in 1982 is unknown.

Experiments were performed to determine the effects of planting seed containing known levels of SMV antigen on antigen content of harvested seeds. Mean ratios (from four, six, and five plots in 1980, 1981, and 1982, respectively) of virus antigen content, as determined by SPRIA, of seeds used to plant the experimental plots compared with antigen content of seeds harvested from plots were 1.04, 3.38, and 0.23 for 1980, 1981, and 1982, respectively.

DISCUSSION

Results of these studies indicated that, on average, plots that had a higher percentage of plants inoculated had a higher percentage of plants infected at time of harvest. The data also show that the highest final disease incidence, and therefore the greatest amount of secondary spread, occurred in 1981 (Tables 1 and 2). This is supported by the observation, noted previously, that SMV antigen content and seed-coat mottling were greatest in seeds from the 1981 harvest. It is also reasonable to assume that the greatest amount of spread would be detected in plots with lower initial levels of disease. This was shown in 1981 and 1982, when the greatest deviation from the expected number of infected plants, assuming no virus spread, occurred in plots where 30% of the plants were initially inoculated (Table 2). In 1983, little apparent secondary spread occurred. Yield losses, which averaged 8% at both the 30 and 50% inoculation

Table 3. Chi-square values in heterogeneity test for spread of soybean mosaic virus in plots where 30 or 50% of the plants were randomly inoculated in 1981-1983

Percentage of plants inoculated	Year and location					
	1981		1982		1983	
	A ^a	B	A	B	A	B
30	6.54	3.26	49.65*** ^b	32.10	4.13	3.26
50	2.86	5.60	4.35	8.09	6.45	8.93

^a Plot A was bordered by woods to the south and grassy pasture on the other sides. Plot B was surrounded by large soybean acreages.

^b Significant at $P < 0.01$.

levels, presumably reflect alterations in competitive ability of soybean plants as described by Ross (14). Previous investigations suggest that yield reductions caused by infection with SMV can be greatest in soybean plants inoculated at early growth stages (3,12). Inoculation of plants as a result of secondary spread probably occurred at different times during the growing season. Yield of plants inoculated at later growth stages probably was not reduced significantly. These data suggest that, at least for the disease caused by SMV in soybeans, secondary spread is more adequately reflected by virus antigen content of seeds than by measurement of yield loss in infected plots.

Experiments to determine the effects of planting seed containing known levels of SMV antigen showed that virus antigen content of seeds harvested from such plots decreased in 1982 compared with the antigen content of seeds used to plant the experimental plots. In 1981, however, the increase of SMV antigen was about threefold. This again agrees with previous data showing that the greatest amount of secondary spread occurred in 1981. The data demonstrate that caution should be exercised by soybean growers who use soybean seed obtained from production fields that may be infected with SMV. Because infection before the onset of flowering results in higher levels of seed transmission (1), the magnitude of potential change in virus antigen content is dependent on the degree and time of secondary spread. Inasmuch as not all seed on an infected plant is infected (1,8), the percentage of SMV-infected seed will gradually decrease when secondary spread is minimal or if spread occurs after the onset of flowering.

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