Using Mixed Cropping to Limit Seed Mottling Induced by Soybean Mosaic Virus

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ABSTRACT

Bottenberg, H., and Irwin, M. E. 1992. Using mixed cropping to limit seed mottling induced by soybean mosaic virus. Plant Dis. 76:304-306.

The spread of aphid-borne soybean mosaic potyvirus (SMV) from inoculated sources was studied in monocultures of soybean and additive mixtures with dwarf (cv. G522DR) or tall (cv. 102F) sorghum. SMV-induced seed mottling, averaged over rows outside the inoculated area, was 4.01% in the soybean monoculture, compared with 2.02 and 2.07% in plots with dwarf and tall sorghum, respectively. Seed mottling decreased rapidly with increasing distance from the inoculum source. Gradients were similar in different treatments, but the intercept was highest in the soybean monoculture. Mean soybean yield per plant was most severely reduced in the tall sorghum mixture, presumably due to competition effects. To minimize competition and protect soybean from aphid-borne virus spread, legumes might, in some circumstances, best be intercropped with cereals of similar height.

A lower incidence of legume viruses has been reported in various legume-cereal mixtures than in legume monocultures (5,11,13,16). Mixed cropping therefore has potential as a low-input cultural practice to protect legumes from aphid-borne virus diseases (15,17). However, the role of canopy structure on aphid-borne virus spread is little understood (8). Knowledge of this relationship is important in devising crop mixtures that offer optimal protection from aphid-borne virus epidemics. Soybean mosaic virus (SMV), a nonpersistently transmitted, aphid-borne potyvirus, affects soybean production worldwide (9). In this study we report the incidence of SMV-induced seed mottling (1,10) in soybean monocultures and mixtures with dwarf or tall sorghum isolines. The main objectives were to determine if the incidence of SMV-induced seed mottling is affected by mixed cropping and if dwarf and tall sorghum isolines affect SMV-induced seed mottling equally. Although the study also helped to elucidate the impact of mixed cropping on soybean yield, it was not designed to identify a cropping system with optimal soybean and sorghum yield for use by farmers in Illinois.

MATERIALS AND METHODS

Treatments. Three types of plantings—monoculures of dwarf sorghum (cv. G522DR), tall sorghum (cv. 102F), and soybean (cv. Williams 82); soybean and dwarf sorghum mixture; and soybean and tall sorghum mixture—were planted on 7 June 1988 at the University of Illinois farm, Urbana. Plantings were planted in paired plots, measuring 5 × 10 m each. Within each pair, one plot was treated with the aphicide Pirimor 50W to control sorghum-infesting colonies of *Rhopalosiphum maidis* (Fitch), a vector of SMV (7). The aphidicide was sprayed into sorghum whorls at 36, 43, 51, and 61 days after planting. The other plot of the pair was not treated. There were four replications. The efficacy of insecticide treatment was determined by counting apterae and alatae in aphid colonies on two sorghum plants per plot at 45 and 52 days after planting.

Plot design. Each plot had 16 soybean rows 0.76 m apart. In mixtures, nine rows of sorghum were planted 1.52 m apart and alternated with pairs of soybean rows. The first and ninth sorghum rows bordered the plots on the west and east side, respectively; the second to the eighth sorghum rows were planted in between two soybean rows each. The distance between a soybean row and the nearest sorghum row was therefore 0.38 m. All rows were 5 m long and oriented east-west. Soybean and sorghum rows were thinned to six and three plants per meter, respectively. To minimize virus spread between plots, 5-m-wide alleyways of oats (cv. Ogle), a nonhost of SMV, were drilled between the plots 9 days after planting. All plots were kept free of weeds during the experiment.

Inoculation of virus source plants. Infected leaves were harvested from 24-day-old soybean plants (cv. Williams 82) that were inoculated 14 days earlier with the SMV-G5 isolate (4). SMV inoculum was prepared by macerating the leaves in a blender with sodium phosphate buffer ($NaPO_4$, 0.01 M) at a weight ratio of 1:5. The suspension was mixed with about 0.6 g of 400-mesh Carborundum powder per liter of inoculum. Six soybean plants in the central 1-m row sections in each of the two center soybean rows were mechanically inoculated 29 days after planting. A cotton wick soaked in the inoculum suspension was rubbed on two leaflets of the youngest fully expanded trifoliolate leaf. To ensure that SMV spread would only initiate from the central inoculum sources, soybean seedlings outside the inoculated area showing seedborne SMV symptoms (severe stunting and chlorosis) were removed immediately after germination. It was assumed from previous experience that immigrant aphids did not carry SMV with them (8).

Determining the percentage of SMV-induced seed mottling and soybean yield. The percentage of SMV-induced seed mottling (7,10) was determined from a 100-seed sample of bulk-harvested seed from 1-m central sections of each of the 16 rows per plot (each section containing five or six plants). Because of labor and time constraints, harvesting and processing of individual soybean plants was not feasible. Mottling was characterized by distinct black saddle-shaped hilum "bleeding" covering at least 10% of the seed coat. Discolorations of questionable nature covering less than 10% of the seed coat were not considered to have been induced by SMV. Soybean seed yield per plant (adjusted to 13% moisture content) was determined by dividing the total seed weight obtained from the 1-m row section by the number of plants in that section. The 100-seed weight was determined from the sample used for assessing seed mottling. Because field space was limited, virus-free control plots were not planted. The impact of SMV infection on soybean yield could therefore not be assessed. All plots (untreated and treated with aphidicide) were sampled for SMV seed mottling and soybean seed yield assessment. Sorghum yield was not determined.

Statistics. To stabilize the error variance and maintain a normal error distribution, the percentage of seed mottling was transformed by using the double square root transformation (3). Mean separation tests (LSD) were carried out only when the ANOVA indicated a significant treatment effect. All analyses were done by using SAS procedures (12).

RESULTS

Aphid colony suppression. Two days after the second aphicide application,

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Accepted for publication 24 October 1991 (submitted for electronic processing).

densities of *R. maidis* apterae on sorghum plants (mean number per plant ± SE, 3 df) in the treated mixtures (23.6 ± 19.7) were lower but not statistically different ($F = 3.07$, $P = 0.18$) from those in the untreated mixtures (65.5 ± 18.0). Alatae densities (1.7 ± 0.7 in the treated plots vs. 3.6 ± 1.3 in the control) showed the same trend ($F = 8.77$, $P = 0.06$). Seven days later (52 days after planting), one day after the third aphicide application, differences in apterae ($F = 21.74$, $P = 0.02$) and alatae ($F = 12.74$, $P = 0.04$) densities between treated and untreated mixtures were significant. At this time, there were 90.8 ± 14.4 apterae and 2.1 ± 0.6 alatae per plant in the controls, whereas the treated plots had 11.3 ± 9.8 apterae and 0.3 ± 0.3 alatae per plant.

Seed mottling. Averaged over all rows outside the inoculated area in treated and untreated plots, soybean monocultures had double the percentage of SMV-induced seed mottling that was found in the dwarf or tall sorghum mixtures (Table 1). Suppression of *R. maidis* colonies did not significantly influence SMV-induced seed mottling in the crop mixtures ($F = 0.48$, $P > 0.5$), soybean seed yield per plant ($F = 0.21$, $P > 0.05$), or the 100-seed weight ($F = 0.10$, $P > 0.05$).

Mottling in the two central inoculated rows averaged 37% in the soybean monoculture, 45% in the dwarf sorghum mixture, and 49% in the tall sorghum mixture but did not differ significantly between treatments ($F = 5.15$, $P > 0.05$). The percentage of seed mottling in individual rows outside the inoculated area decreased sharply with increasing distance from the center (Fig. 1). Regression analysis showed that the slopes of the gradients did not differ significantly between treatments ($F = 0.69$, $P > 0.05$) (Fig. 2). However, the soybean monoculture had a significantly higher intercept than either the dwarf sorghum or tall sorghum mixture ($F = 5.22$, $P < 0.05$, LSD = 0.77).

Soybean yield. Averaged over all rows outside the inoculum area in insecticide-treated and untreated plots, total seed weight per plant (gram) was greatest in the soybean monoculture, least in the tall sorghum mixture, and intermediate in the dwarf sorghum mixture (Table 1). The 100-seed weight did not differ significantly among treatments.

On a per-row basis, soybean yield per plant increased slightly with increasing distance from the inoculated rows. This trend was most obvious in the soybean monoculture but seemed absent in the crop mixtures where rows with high yields alternated with rows of low yields (Fig. 1). The 100-seed weight varied little across rows. In the soybean monoculture, soybean yield per plant was linearly and negatively correlated with the percentage of seed mottling ($r = -0.95$, $P < 0.001$). No correlation between yield and seed mottling was found in the dwarf sorghum ($r = -0.32$, $P > 0.05$) or tall sorghum mixtures ($r = +0.74$, $P > 0.05$).

DISCUSSION

Seed mottling can be regarded as an indication that the parent plant was infected with SMV (8). In this study, the percentage of seed mottling was assessed from the combined seed yield of six plants in a row sample. The major factor affecting the percentage of seed mottling therefore is the number of plants in each sample that became infected and produced mottled seeds. Soybean plants in the center rows, which were inoculated 29 days after planting, produced 37–49% seed mottling, whereas plants in rows next to the center rows developed no more than 12% mottling. However, all plants in the 1-m row section next to the inoculated center rows eventually showed mosaic symptoms typical of SMV (H. Bottenberg, *unpublished data*). This suggests that the percentage of seed mottling decreased with increasing plant age at the time of infection, but the mode of infection (early mechanical inoculation vs. later transmission by aphids) may also have played a role. Other researchers were unable to detect a relationship between seed mottling and time of infection (1).

Table 1. Percentage of seed mottling induced by soybean mosaic virus[y], soybean seed weight per plant, and 100-seed weight in soybean monocultures and in soybean-sorghum mixtures (averaged over all uninoculated rows). Means (± SE, 3 df) within columns followed by the same letter are not significantly different as determined by LSD mean separation ($P < 0.05$)

Treatment	SMV-induced seed mottling (%)	Seed weight per plant (g)	100-seed weight (g)
Soybean monoculture	4.01 ± 0.61 a	20.54 ± 1.35 a	15.54 ± 0.26
Dwarf sorghum and soybean	2.02 ± 0.42 b	15.38 ± 1.34 b	15.43 ± 0.27
Tall sorghum and soybean	2.07 ± 0.50 b	9.83 ± 0.83 c	14.69 ± 0.23
F value	22.01**	65.12**	3.71[z]

[y]Untransformed data shown, analysis performed on double square root transformed data.
[z]Not significant, ** $P < 0.01$.

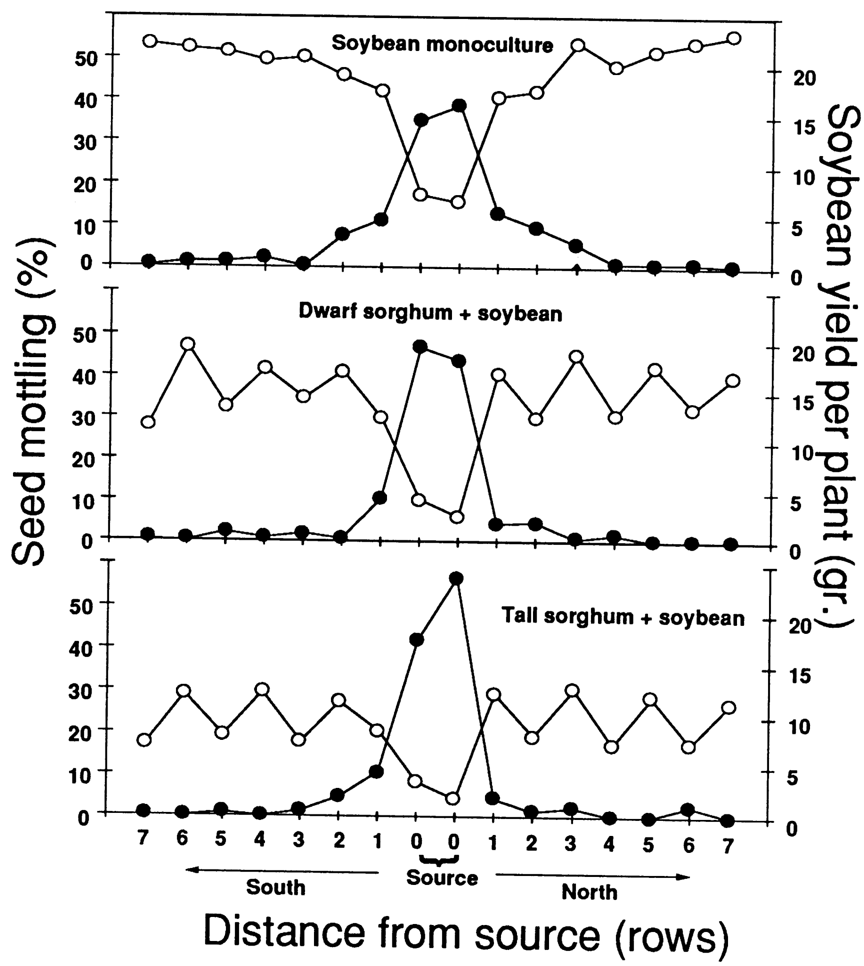

Fig. 1. Percentage of seed mottling (●) and soybean yield per plant (○) in soybean monocultures and soybean-sorghum mixtures.

Mixed cropping significantly reduced the percentage of SMV-induced seed mottling. This reduction may be related to lower aphid landing rates and/or the higher probability of aphids losing infectivity after landing and probing on sorghum, a nonhost of SMV, in crop mixtures (6). Dwarf and tall sorghum reduced seed mottling equally, indicating that sorghum height was not a major factor in limiting the spread of SMV.

The role of local, sorghum-infesting *R. maidis* colonies in SMV spread could not be assessed because colony suppression in insecticide-treated plots may not have been successful. Densities of alatae and apterae were significant only after the third insecticide treatment. More frequent insecticide applications would be required to prevent aphid colonies on sorghum and, consequently, to accurately assess the importance of local aphid colonies in SMV spread. Also, colony counts should be made more frequently, at least one after each insecticide application. Despite incomplete aphid colony suppression in this study, however, it seems logical to assume that such colonies in the untreated plots did not contribute to virus spread because, if they would have, SMV seed mottling would have been greater in the untreated sorghum-soybean mixtures.

The gradient in seed mottling may be regarded as a reflection of the movement pattern of viruliferous aphids that transmitted the virus from the central source to surrounding plants. The greater intercept of the seed mottling gradient in the soybean monoculture indicates that more vectors may have landed in this habitat than in the mixtures. However, the similar slopes of the gradients indicate that the rate of vector movement across rows may have been similar in both soybean monocultures and crop mixtures. Sorghum plants may not have presented a major barrier to

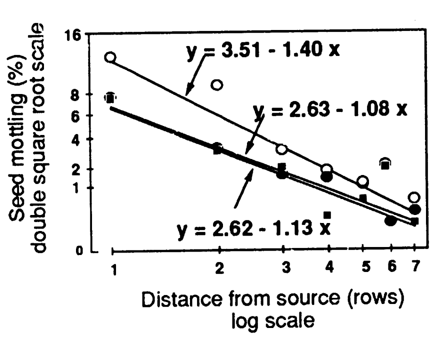

Fig. 2. Linear regression of percentage of seed mottling induced by soybean mosaic virus (y, double square root transformed) at different distances from a central inoculum source (x, natural log transformed) (averaged over directions) in soybean monocultures (○), dwarf sorghum-soybean mixtures (●), and tall sorghum-soybean mixtures (■). Equations shown are based on transformed values.

vector movement because numerous gaps in the mesh of sorghum foliage (H. Bottenberg, *unpublished data*) allowed aphids to fly freely across rows.

Despite the higher percentage of seed mottling, soybean yield was highest in the soybean monoculture. Interplant competition might have been a greater constraint to soybean yield than SMV infection, probably because the central inoculum source only comprised 2.5% of the total soybean plant population. Because virus-free controls were not available, an accurate comparison of the relative impact of mixed cropping and SMV infection on soybean yield is not possible.

Shading by tall and dwarf sorghum was apparently responsible for reducing soybean yield by an average of 25 and 50%, respectively. Competition for nutrients and moisture by sorghum may also have been involved to a limited extent but would not explain the difference observed between the tall and dwarf sorghum-soybean mixtures. Soybean rows that were directly north of a sorghum row were shaded more and yielded less than other soybean rows, causing a jagged yield gradient across the field (Fig. 1). Lower seed weight per plant was related to a lower production of seeds because the 100-seed weight varied little across rows and treatments. Likewise, Thompson et al (14) found significantly fewer soybean pods when soybean was grown with tall maize than when grown with dwarf maize.

Because sorghum yield was not determined in this study, it is not possible to quantify total biomass and economic return of dense and sparse sorghum-soybean mixtures. It would be difficult therefore to give practical recommendations to farmers. However, it seems plausible that, under certain conditions, farmers would benefit most by intercropping soybean with dwarf sorghum, which would protect the soybean from aphid-borne virus spread while minimizing yield loss due to shading by sorghum. Mixed cropping is most practical and economical under low-input, small-scale farming conditions, mostly in developing countries. Because mixed cropping, in its conventional row-by-row arrangement, requires intensive manual care from planting to harvesting, it is not suited for large-scale, mechanized agriculture. Strip intercropping, a form of mixed cropping in which different crops are planted in strips of multiple rows, is better adapted to mechanized farming and has been shown to suppress certain pest populations (2). However, its impact on aphid-borne virus spread is difficult to predict and would require further study.

ACKNOWLEDGMENTS

We thank Doyle Dazey for planting the field; Gail Kampmeier for technical advice; Ellen Brewer and Joan Alster for statistical advice; and Gail Kampmeier, David Onstad, William Ruesink, and the Illinois Natural History Survey editorial staff for reviewing earlier versions of this paper. This work was carried out as part of an entomology Ph.D. dissertation by the first author and was supported by USAID Program Support Grant DPE-5542-G-SS-6013-00.

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