Evaluation of Sorghum Hybrid Mixtures for Controlling Sorghum Leaf Blight

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ABSTRACT

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This is the first report on the use of mixtures of sorghum genotypes to control a major pathogen. The effect of mixtures of sorghum hybrids on development of sorghum leaf blight caused by *Exserohilum turcicum* was studied in three locations in central Mexico. Seed of resistant and susceptible sorghum hybrids were mixed in resistant:susceptible ratios of 1:0, 3:1, 1:1, 1:3, and 0:1. Some disease developed on the resistant hybrid, but disease severity varied little on the resistant component of mixtures in any location. Only in one location was disease severity on the resistant hybrid observed to significantly increase with increasing proportion in the mixture ($P < 0.05$). This increase, however, was more than compensated for by reduction in disease severity in the susceptible hybrid. In contrast, there were significant differences in the amount of diseased tissue on the susceptible hybrid ($P < 0.05$) as the percentage of the resistant component was increased in all locations. Although disease amelioration was more noticeable in locations with higher disease pressure, it was significantly greater under conditions of low disease incidence. Every addition of 1% of resistant plants to the susceptible plant population significantly reduced disease development by up to 0.4% on the susceptible hybrid ($P < 0.05$). Furthermore, mixtures had a significant effect on yield of the susceptible hybrid but not on yield of the resistant hybrid. Adding 1% of resistant plants to the mixture increased yield of the susceptible plant by up to 0.55%.

Additional keywords: disease management, gene deployment, northern corn leaf blight

Exserohilum turcicum (Pass.) K.J. Leonard & E.G. Suggs, the causal agent of leaf blight on sorghum (*Sorghum bicolor* (L.) Moench) and maize (*Zea mays* L.), is an economically important pathogen in the United States, Argentina, Mexico, and Israel (7). Sorghum grain losses caused by this disease are usually low (6) but can be severe in susceptible sorghum cultivars if the disease is established before panicle emergence (1; W. de Milliano, *personal communication*).

The El Bajio area of central Mexico is the second largest sorghum production area of the country, with over 0.75 million hectares planted to this cereal. In this region, leaf blight is considered to be a limiting factor for sorghum production (2). Although the most practical method of control is host plant resistance, most commercial hybrids used in that area are moderately susceptible. No blight-resistant hybrid with yield potential equal to the currently planted hybrids is available to replace them.

Mixing resistant and susceptible plants in the same field is one method proposed to reduce disease severity on susceptible plants (3,5,10,17,20). The mixture may consist of inbred lines, hybrids, cultivars, or clones. However, hybrid mixtures provide the same disease protection with the advantage over other mixture types of higher yield potential. Genetic diversification of host populations for disease resistance is expected to stabilize pathogen populations, thereby preventing the breakdown of new disease resistance (5). Mixtures have been effective in reducing the severity of crown and stem rust on oats (5,12); powdery mildew on wheat, barley, and oats (20); and leaf blast on rice (11), among others. There is little information on the value of host diversity on sorghum cropping. The promise of mixtures for not only increasing but also stabilizing yields is important for the millions of people in developing countries who depend on this cereal as their main staple food. The objective of this work was to determine the effect of mixtures of resistant and susceptible hybrids on blight severity and on crop yield.

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MATERIALS AND METHODS

Three trials were conducted in Mexico in 1987 using two hybrids that differed in leaf blight susceptibility. Two of the trial sites, Ocotlan and Salamea, had been characterized by severe blight damage to sorghum in previous years. The experimental design at Ocotlan and Salamea was a randomized complete block with five treatments and eight replications. The treatments were plantings of five mixtures of seed of two hybrids. Ratings for the resistant Purepecha and susceptible RB104x25 hybrids varied between 1.5 and 2.5 (based on a 1–5 scale, with 1 being completely resistant and 5 being completely susceptible) and between 3.5 and 4.5, respectively, during the 3 yr prior to this experiment at these locations. The seed mixtures were resistant:susceptible ratios of 1:0, 3:1, 1:1, 1:3, and 0:1. Each experimental plot consisted of 10 8-m rows spaced 76 cm apart. Maize barriers 2.4–3 m wide were planted between adjacent plots to decrease spore movement. Maize is not a host for isolates of *E. turcicum* attacking sorghum. A maize barrier 3 m wide also surrounded the whole experiment. A third trial was planted at Celaya, where leaf blight had been less severe. Each experimental plot there consisted of three 5-m rows spaced 76 cm apart with no maize barriers between them.

The components in the mixtures were chosen because of their similarity in maturity and yield and their differences in easily identifiable characters such as midrib characteristics, grain color, and plant color (in response to injury). These differences made it possible to identify the genotype of each plant when taking disease notes. Seeds were mixed manually before planting. The number of seeds of each component in the mixture was adjusted by the percentage of seed germination. Initial inoculum probably came from the large commercial sorghum plantings within which each experiment was located.

Percent leaf area affected by blight was measured on each of the upper four to six leaves. Disease scorings in Ocotlan roughly coincided with flag leaf emergence, panicle emergence, milk stage, and dough stage of grain; in Salamea with flag leaf emergence, panicle emergence, and dough stage; and in Celaya with the flowering and dough stages. The total number of plants sampled per plot varied from 20 to 25 at the first and second readings and from 50 to 75 at the final two readings. The number of susceptible plants sampled per plot was twice the number of resistant plants sampled on each date to compensate for their larger variability. Plants were sampled randomly from the central 6 m of each of the middle eight rows of each plot in Ocotlan and Salamea and from every row at Celaya. The scores for each genotype were kept separate. The number of resistant and susceptible plants in each plot was assessed 2 wk before harvesting. These numbers, in general, did not deviate by more than 4% from expected values. Data for resistant and susceptible plants were analyzed separately. The corresponding area under the disease progress curve (AUDPC) for the three upper leaves of susceptible plants was calculated using a FORTRAN program written by W. Schuh (Department of Plant Pathology, Penn State) and C. Brown (Department of Plant Pathology and Microbiology, Texas A&M). To test the effect of mixtures on disease severity of susceptible plants throughout the growing season, AUDPC values were regressed on the proportion of susceptible plants in the mixtures. To test whether the proportion of resistant plants in the mixture had an effect on disease severity of either the susceptible or the resistant hybrid at different growth stages, the regression of disease severity against percentage of resistant plants in the mixture was calculated and the null hypothesis of slope *b* being equal to 0 was tested following standard statistical methods (9).

The middle four rows of each plot in Ocotlan and whole plots in Celaya were harvested. Heads from each hybrid were separately threshed and weighed, and their yield was adjusted to 12% moisture content of the grain. The yield of each genotype was adjusted by dividing by 4, 3, 2, or 1 according to its frequency (100, 75, 50, or 25%) in the mixtures. Yield data from Salamea was not obtained because of bird damage. The adjusted yield of each hybrid was regressed against its proportion in the mixtures.

RESULTS

In Ocotlan, disease was first observed 30 days after planting (six-leaf stage). At flowering, only lower leaves of the susceptible hybrid were severely damaged, but at maturity, even the flag leaf of this hybrid was severely diseased (Fig. 1). Disease amelioration in the mixtures was considerable. The disease progress curve for the flag leaf of the susceptible component was considerably lower in the mixtures. The AUDPC for the three upper leaves of the susceptible hybrid decreased when the proportion of resistant plants in the mixture increased (Fig. 2). Linear effects were significant ($P < 0.05$) for the AUDPC values for two of the three upper leaves in this location.

In Salamea, disease severity on the susceptible hybrid was less affected by the mixtures than in Ocotlan. The flag leaves of susceptible plants had up to 30% less disease severity in the mixtures at dough stage (Fig. 3). Disease amelioration was similar for the mean of the upper three leaves. The AUDPC values for the mean of the three upper leaves of susceptible plants were reduced when the

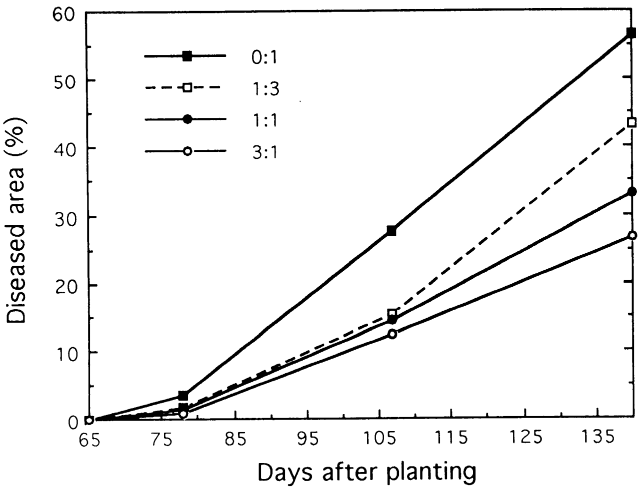

Fig. 1. Disease progress on the flag leaf of the susceptible hybrid RB104x25 planted in mixtures with the resistant hybrid Purepecha or as pure stands at the Ocotlan site. Legend shows the ratio of resistant to susceptible plants.

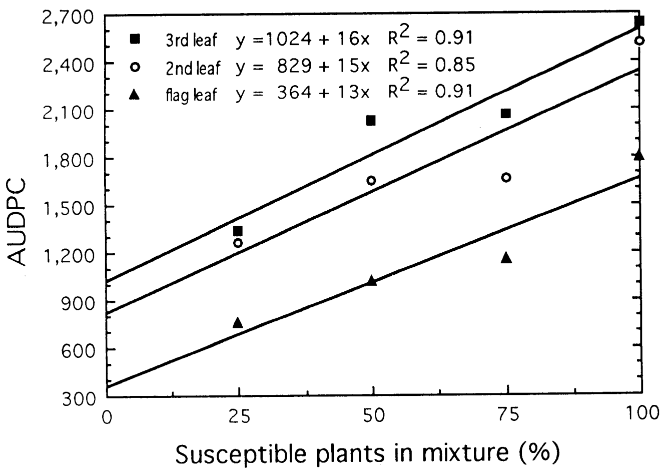

Fig. 2. Effect of the proportion of susceptible plants in the mixture on the area under the disease progress curve (AUDPC) of the susceptible hybrid RB104x25 planted at the Ocotlan site. In the equations, *y* is disease severity on the susceptible plant and *x* is percentage of susceptible plants in the mixture. All but the second of the coefficients of determination (r^2) are significant at $P < 0.05$.

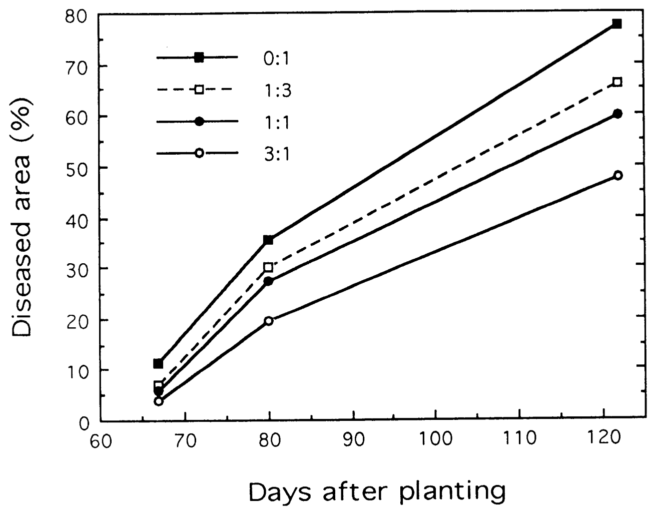

Fig. 3. Disease progress on the flag leaf of the susceptible hybrid RB104x25 planted in mixtures with the resistant hybrid Purepecha or as pure stands at the Salamea site. Legend shows the ratio of resistant to susceptible plants.

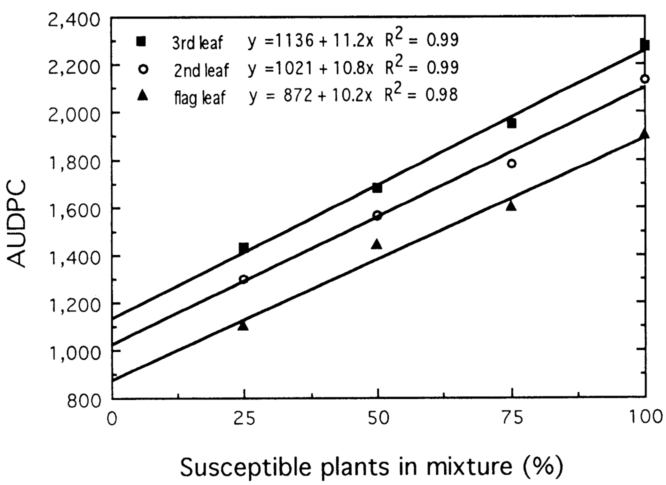

Fig. 4. Effect of the proportion of susceptible plants in the mixture on the area under the disease progress curve (AUDPC) of the susceptible hybrid RB104x25 planted at the Salamea site. In the equations, *y* is disease severity on susceptible plants in the mixture. All coefficients of determination (r^2) are significant at $P < 0.01$.

number of resistant plants in the mixtures increased (Fig. 4). Linear effects were significant for the mean of the three upper leaves.

Celaya was the location with the lowest disease severity. The highest percentage of diseased area was always observed on the susceptible hybrid planted in pure stands and varied from 15% in the flag leaf to 18% in leaf 5. Disease reduction on the susceptible hybrid planted in mixtures in this location was lower than in the other locations but still significant for leaves 1, 2, and 4 at the dough stage (Table 1).

Slopes for the regression lines of disease severity on the susceptible hybrid against percentage of resistant plants in the mixture were significantly different from zero at the dough stage in all locations (Table 1). In Salamea, the mixture effects were consistently significant as early as panicle emergence, while in Ocotlan and Celaya, significant mixture effects were not noticed until dough stage. In contrast, the slopes for disease severity on the resistant hybrid against the percentage of susceptible plants in the mixture were significantly different from zero only in Ocotlan (Table 2). In this location, leaves 2, 3, and 4 of the resistant hybrid showed a slight increase of disease severity when the proportion of susceptible plants was increased in the mixture. This increase, however, was more than compensated for by a larger decrease of disease severity on the susceptible hybrid in the same mixtures (Table 1). Sorghum leaf blight on the susceptible hybrid decreased in all locations as the number of resistant plants in the mixture increased, whereas the disease on the resistant hybrid slightly increased in Ocotlan as the number of susceptible plants in the mixture increased (Table 2).

In Ocotlan, the susceptible hybrid yielded significantly more when planted in mixtures than when planted alone (Fig. 5). The regression line predicted a yield increase of 17.8 kg/ha for the susceptible hybrid per each 1% addition of the resistant hybrid to the mixture ($P = 0.025$). In contrast, yield of the resistant hybrid was not significantly affected by the mixtures ($P = 0.25$) (Fig. 5).

In Celaya, mixtures had no significant effect on yield of either the resistant or the susceptible hybrid (Fig. 6). However, the P value for the slope of the regression line for the susceptible hybrid was nearly significant ($P = 0.06$). The value of the slope for the susceptible hybrid was very similar in Ocotlan and Celaya.

DISCUSSION

Little research has been done on the use of sorghum mixtures to control diseases. Only one other report (18) mentions the testing of sorghum hybrid mixtures, and these were obtained under varied and often stressed growth conditions. In that study, mixtures did not perform better than pure stands, but disease was apparently not a limiting factor. In our experiments, adding resistant plants to the susceptible plant population significantly reduced disease severity. The regression of AUDPC values vs. percentage of resistant plants at Ocotlan indicates that adding 25, 50, and 75% resistant plants to the mixture results in approximately 20, 40, and 60% lower AUDPC for flag leaves of the susceptible component, respectively. In Salamea, where the disease was more severe, the predicted AUDPC reduction for flag leaves was approximately 14, 27, and 40%, respectively. It is interesting that when disease pressure was higher, there was less disease reduction in mixtures.

Mundt and Leonard tested resistant and susceptible mixtures of oat plants (13) and maize hybrids (15). They reported that the effectiveness of mixtures for disease control decreased with increasing genotype unit area and with increasing steepness of the dispersal gradient. The area occupied by individual plants in this sorghum crop (about 0.055 m^2) is somewhat large compared to the area for individual plants of crops such as wheat (0.003 m^2) where the benefits of mixtures have been demonstrated. Mundt and Leonard (14) reported that when the initial inoculum is uniformly distributed over the plot (as it was in this study), the effectiveness of mixtures of plants with large host genotype unit area in reducing disease is greatly decreased. In our case, the host genotype unit area was never as large as the one they used (0.25 m^2 vs. 0.055 m^2). Host genotype unit area could still be smaller for the region, since farmers use higher plant densities and, in theory, can realize more benefits from mixtures.

Leaf blight resistance on the sorghum hybrid Purepecha has been observed as fewer lesions, shorter incubation period, and lower production of spores per lesion as compared with the susceptible hybrid RB104x25 (19). Increased inoculum pressure tends to produce more lesions on both hybrids. However, *E. turcicum* has about 50% lower infection efficiency in Purepecha than in the susceptible hybrid (19). This can explain the fact that although the number of lesions on the resistant hybrid increased in mixtures, disease severity on the resistant hybrid remained low in all locations.

The disease amelioration observed in

Table 1. Coefficients of regression of percent disease severity on the upper leaves of susceptible plants vs. percentage of resistant plants in mixtures of resistant and susceptible sorghum hybrids[a]

Location Growth stage	Leaf 1	Leaf 2	Leaf 3	Leaf 4	Leaf 5
Ocotlan					
Flag leaf	0.00[b]	0.00	−0.03	−0.09*	−0.11*
Panicle	−0.03	−0.04	−0.06	−0.10*	−0.19
Milk	−0.18	−0.18	−0.19	−0.18	−0.24
Dough	−0.40*	−0.36*	−0.34*	−0.27*	−0.14
Salamea					
Flag leaf	−0.10*	−0.13	−0.17*	−0.20**	−0.22*
Panicle	−0.20*	−0.25**	−0.30**	−0.29**	−0.26*
Dough	−0.38**	−0.34**	−0.30**	−0.21**	−0.12*
Celaya					
Flowering	−0.02	−0.03	−0.01	−0.03	−0.01
Dough	−0.11**	−0.09*	−0.04	−0.12*	−0.04

[a] Disease was measured during different growth stages in locations of central Mexico during 1987. Leaves are numbered from the top down.
[b] * = Significant at $P < 0.05$, ** = significant at $P < 0.01$.

Table 2. Coefficients of regression of percent disease severity on the upper leaves of resistant plants vs. percentage of susceptible plants in mixtures of resistant and susceptible sorghum hybrids[a]

Location Growth stage	Leaf 1	Leaf 2	Leaf 3	Leaf 4	Leaf 5
Ocotlan					
Flag leaf	0.00[b]	0.00	0.01	0.01	0.01
Panicle	0.00	0.00	0.01	0.05	0.00
Milk	0.00	0.00	0.00	0.01	0.01
Dough	0.01	0.02*	0.03**	0.02*	0.02
Salamea					
Flag leaf	0.01	0.01	0.03	0.03	0.01
Panicle	0.00	0.00	0.00	0.00	0.01
Dough	0.02	0.02	0.02	0.03	0.04
Celaya					
Flowering	0.00	0.01	0.00	0.00	0.00
Dough	0.01	0.03	0.01	0.02	0.01

[a] Disease was measured during different growth stages in locations of central Mexico during 1987. Leaves are numbered from the top down.
[b] * = Significant at $P < 0.05$, ** = significant at $P < 0.01$.

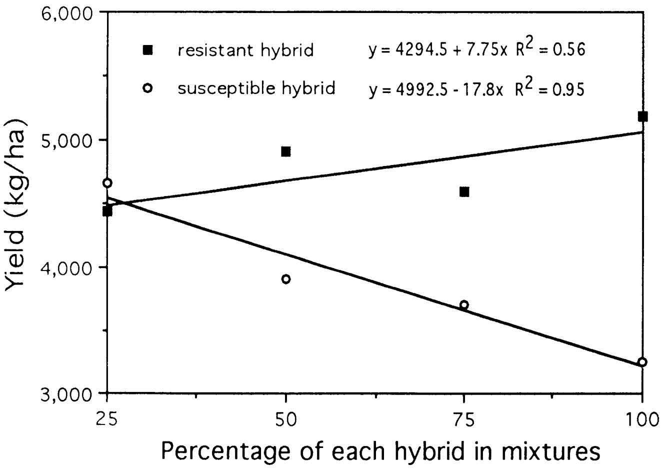

Fig. 5. Yields of susceptible and resistant plants in relation to their proportions in mixtures of resistant and susceptible sorghum hybrids planted at the Ocotlan site.

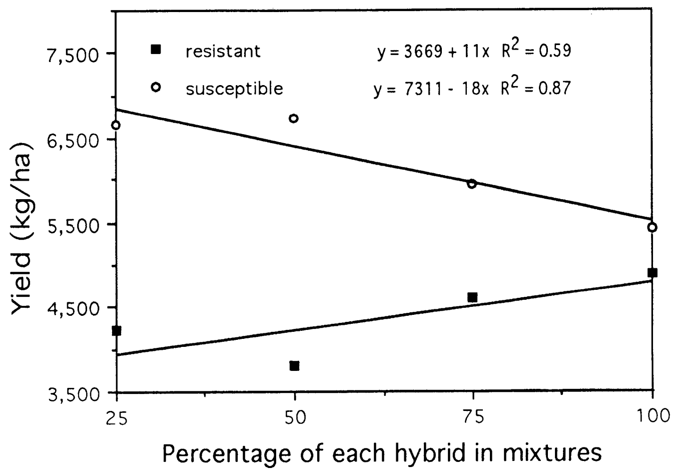

Fig. 6. Yields of susceptible and resistant plants in relation to their proportions in mixtures of resistant and susceptible sorghum hybrids planted at the Celaya site.

the mixtures translated to higher yields in the susceptible hybrid in Ocotlan. Even under the conditions of low disease pressure found in Celaya, the tendency of yield increase for the susceptible hybrid in mixtures approached the arbitrary 0.05 significance level ($P = 0.06$). In contrast, yield of the resistant hybrid was not significantly affected by the mixtures in either location. Regression lines predicted that adding 25, 50, and 75% of resistant plants to the mixture would translate to 13.8, 27.6, and 41.4% increase in the yield of the susceptible hybrid, respectively. This shows not only the capacity of *E. turcicum* to cause plant damage but also that mixtures can effectively protect the crop under a wide range of disease pressure.

In a study of yield loss caused by *E. turcicum* on maize, substantial loss of grain yield was associated with high levels of diseased leaf area (16). In our studies, the pathogen damaged over 55% of the flag leaf of the susceptible hybrid in Ocotlan and over 75% in Salamea; therefore, some yield loss was expected. However, there is a need to establish the yield loss due to different levels of leaf blight damage on sorghum to better characterize these effects. Data reported here might also be applied to maize, since the plant is similar in size to sorghum and the pathogen is important in both crops.

One advantage of cultivar mixtures is that they allow the use of a resistant but lower yielding cultivar to protect a susceptible but higher yielding cultivar from disease in the mixture. Therefore, it is important to determine what proportion of resistant plants is needed to minimize yield losses for the susceptible cultivar. Browning and McDaniel (cited in 4) determined that 33% of specific pathogen resistance was adequate to protect an oat population against crown rust even in an environment favorable for disease development. The levels of resistance needed to protect a plant population may vary in different environments (5). In order to keep a safe margin, researchers advocated use of at least 60% of the plants resistant to each of the prevalent races of the targeted pathogen population (8). In our study, both hybrids had similar yield potential and the mixtures did not yield significantly more than the best pure stand. However, on the basis of disease development in the mixtures, we recommend 50% resistant plants for sorghum cultivar mixtures.

There are several benefits to using sorghum cultivar mixtures. First, the control of diseases such as anthracnose and sorghum downy mildew, which have shown great ability to overcome major resistance genes, may be enhanced. Second, a breeding program specifically for mixture development is not needed, since the strategy allows for the immediate use of high yielding hybrids or cultivars as they become available. Third, components in mixtures could be chosen to control more than one disease and/or pest. If sorghum mixtures are effective in decreasing the rate of spread of several pathogens and/or pests at the same time as expected, it would be easier to convince farmers to use mixtures. Implementation of the strategy is of little cost to the farmer (21). Therefore, mixtures may be suitable for use in developing countries to ensure higher and more stable yields.

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