Specificity of the *Bipolaris sacchari-Saccharum* spp. Interaction

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ABSTRACT

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Nine and eight sugarcane (*Saccharum* spp.) clones were inoculated with four and five isolates of *Bipolaris sacchari*, respectively. Interorganismal genetic techniques were used to study the specificity between *Bipolaris sacchari* and *Saccharum* spp. In both experiments, three corresponding gene pairs for low reaction in sugarcane and low pathogenicity in *B. sacchari* were postulated. Low-high infection type differences resulting from *B. sacchari-Saccharum* spp. interaction indicated specificity (gene-for-gene relationship) between the host and the pathogen.

Additional key words: eyespot, host-parasite interaction

Eyespot of sugarcane (*Saccharum* spp.) incited by *Bipolaris sacchari* (But.) Shoem. has occasionally been of economic importance in the continental United States, especially during the development of the sugarcane industry when eyespot-susceptible "noble" (*Saccharum officinarum* L.) cultivars were grown here (1). Miller (9) evaluated clonal selections in the Florida sugarcane cultivar development program at Canal Point for eyespot resistance and suggested that a screening technique should be applied in the early stages of selection to eliminate large numbers of susceptible clones. Dean and Miller (3) developed a rapid field-screening technique for eyespot resistance, and indicated that eyespot resistance of the screened population was significantly higher than that of the original unscreened population. Although effectiveness of selection for eyespot resistance has been reported, information on specificity of *B. sacchari-Saccharum* spp. interaction is lacking. The concept of interorganismal genetics, as discussed by Loegering (6), has not thus far been applied in the study of sugarcane diseases. The principles of interorganismal genetics make it possible, without making crosses, to determine hypothetical genotypes of the two symbiotes making up an association (8). Gene-for-gene associations have been demonstrated for several nonobligate parasite-host interactions (2), in which discrete infection types could be identified. *B. sacchari* is a nonobligate pathogen.

The objective of these investigations was to study the specificity of the *B. sacchari-Saccharum* spp. interaction by use of the interorganismal genetic techniques as described by Loegering (5–7).

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MATERIALS AND METHODS

1983 Experiment. Nine sugarcane clones (cultivars) from diverse genetic backgrounds (CP 57-603, CP 77-1008, CP 78-1701, CP 78-2100, CP 82-1249, CP 82-1565, CP 82-1720, CP 83-1480, and CP 83-1484) were used in this experiment. All of these clones, except CP 77-1008, had shown eyespot or eyespotlike symptoms in the field. Single-bud seed pieces were planted in plastic pots (17 cm diameter and 18 cm deep) filled with "muck" soil (histosol or organic soil) on 11 November 1983 with the objective of obtaining four viable plants per clone per isolate for inoculation. Four isolates (1701, 1463, 2100, and NP4) of *B. sacchari* were used. Each fungal isolate was initiated from a single conidium and grown under ultraviolet light on lactose casein hydrolysate agar (to promote sporulation) for 14 days. A culture of each isolate was homogenized in a blender, diluted with a 1% molasses solution to a conidial concentration of 50,000/ml, and sprayed on the plants from a mist-generating spray bottle. One plant per clone per isolate was inoculated on each of four consecutive days to give four replicates. It was necessary to replicate in time rather than in space because of the limited space in the dew chamber where the plants were held in a saturated atmosphere for 24 hr after inoculation before they were transferred to the greenhouse.

Plants were rated for infection type as high (H) or low (L) 5 days after the last day of inoculation. Low infection type was characterized by small brown spots with little or no surrounding halo and little or no elongation in the long direction of the leaf blade (lesion length <5 mm). High infection type was characterized by relatively large brown spots and yellow halos that were prominent and elongated in the long direction of the leaf blade, but the halo was elongated more than the brown spot, giving the typical "eyespot" for which the disease is named (lesion length ≥ 10 mm).

The H-L system of hypothesizing corresponding gene pairs in the host and the pathogen was used as elucidated by Loegering (5,7). A gene-for-gene relationship is assumed. In this system, pathogenicity is considered a characteristic of the pathogen and is designated as low (Lp) or high (Hp). For each gene for pathogenicity in the pathogen, there is a corresponding gene for reaction in the host that may be low (Lr) or high (Hr). The host-pathogen genetic combination Lr/Lp results in low infection type and is definitive. The remainder of possible combinations Lr/Hp, Hr/Lp, and Hr/Hp result in high infection type (H). This interaction between corresponding gene pairs has been designated as the category III interaction, and the interaction among category III interactions has been designated as category IV interaction (5).

1984 Experiment. Eight genetically diverse clones of sugarcane (CO 453, CP 65-357, CP 70-1133, CP 78-2100, Hinahina, NG 77-82, NG 77-103, and NG 77-234) and five isolates of *B. sacchari* (1203, 1231, 1247, 1475, and 2100) were used. Unfortunately, all the isolates used in the previous experiment were not available. Single-bud seed pieces of the eight clones were planted in pots (same size and soil type as described previously) on 28 November 1984. Procedures for inoculum preparation and inoculating plants were the same as described for the previous experiment. One plant per clone was inoculated with each isolate. Inoculations were done on 15 January 1985. The inoculated plants were kept in dew chamber for 24 hr, then transferred to a greenhouse. Plants were rated for infection type after 8 days. Corresponding gene pairs in the host and the pathogen were hypothesized using the L-H system.

In both 1983 and 1984 experiments, uninoculated controls were used. The controls showed no symptoms of eyespot.

RESULTS AND DISCUSSION

In the 1983 experiment, the L-H system showed differences among clones in their reactions to the eyespot pathogen (Table 1). Similarly, isolates also differed in their pathogenicity (Table 1). However, isolates 1701 and 1463 showed identical pathogenicities on clones in this study. The L or H designation assigned to the nine clones for each of the four inoculation days (replications) was the same as shown in Table 1. Using the L-H system of Loegering (5,7), we postulated a minimum of three corresponding gene pairs (CGPs) for the clones and isolates used in the 1983 experiment.

In the 1984 experiment also, differences in reaction among clones existed (Table 2). Isolates 2100, 1475, and 1231 were identical in pathogenicity but differed from the other two isolates. An examination of infection types (Table 2) indicated three CGPs for low reaction in the host and low pathogenicity in the pathogen.

It would have been beneficial to have the same isolates in both the 1983 and 1984 experiments, but maintenance of isolates was difficult because of their instability in culture. In both experiments, the L-H differences resulting from the *B. sacchari-Saccharum* spp. interaction indicated specificity or gene-for-gene association. Earlier, Robinson (10,11) indicated that gene-for-gene associations do not occur in certain plants, one of which is sugarcane, because of the conditions under which the plants evolved. The argument is that sugarcane has a polyphyletic origin from ancestral grasses that evolved in a continuous wild pathosystem in which no selection pressure for a gene-for-gene system could have been applied (11). Some of Robinson's assumptions have been questioned (4), but this paper presents the first experimental evidence for the existence of a gene-for-gene relationship in sugarcane and one of its pathogens (*B. sacchari*).

We analyzed the data with the assumption that low infection type is caused by specific host-pathogen interactions. The data could also be analyzed with the assumption that high infection type is caused by specific interactions. Under the latter possibility, three CGPs could be postulated in both experiments. Although the number of CGPs remained the same as under the first assumption, the definitive genes would now reside in different clones and cultures from those postulated under the first assumption.

Table 1. Infection types observed on nine sugarcane clones inoculated with four isolates of *Bipolaris sacchari* (1983 experiment)

	Isolate of *B. sacchari*			
Clone	**1701**	**2100**	**1463**	**NP4**
CP 57-603	H[a]	H	H	H
CP 82-1249	H	L(1)[b]	H	H
CP 78-1701	H	H	H	H
CP 83-1480	H	L	H	H
CP 82-1720	H	L	H	H
CP 82-1565	H	H	H	H
CP 83-1484	H	L	H	L(2)
CP 78-2100	H	H	H	H
CP 77-1008	L(3)	L	L	L

[a] H = high and L = low infection type.
[b] Postulated corresponding gene pairs for low reaction gene(s) in host (Lr) and low pathogenicity gene(s) in pathogen (Lp).

Table 2. Infection types observed on eight sugarcane clones inoculated with five isolates of *Bipolaris sacchari* (1984 experiment)

	Isolate of *B. sacchari*				
Clone	**1247**	**2100**	**1475**	**1203**	**1231**
NG 77-103	H[a]	H	H	H	H
NG 77-82	H	H	H	H	H
CP 78-2100	H	H	H	H	H
CO 453	H	H	H	H	H
CP 70-1133	H	H	H	L(1)[b]	H
CP 65-357	H	H	H	L	H
Hinahina	L(2)	H	H	L	H
NG 77-234	L	L(3)	L	L	L

[a] H = high and L = low infection type.
[b] Postulated corresponding gene pairs for low reaction gene(s) in host (Lr) and low pathogenicity gene(s) in pathogen (Lp).

Sugarcane is a highly polyploid crop that behaves as a diploid (12). However, regular genetic analyses for disease resistance or other traits have been almost nonexistent, if not impossible. Under such circumstances, principles of interorganismal genetics provide sugarcane breeders and pathologists an opportunity to analyze the "disease" data in a more meaningful and logical manner. Because of the polyploid nature of sugarcane, there may be multiple copies of genes for reaction in sugarcane for each corresponding gene for pathogenicity in the pathogen. Because of this possibility of multiple copies in sugarcane, hypothetical genotypes of the two symbiotes as discussed by Loegering and Burton (8) could not be easily determined. However, identification of sugarcane clones possessing genes for low reaction should be useful in a sugarcane breeding program.

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