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## FALL ARMYWORM (LEPIDOPTERA: NOCTUIDAE) RESISTANCE IN TEXAS BLUEGRASS, KENTUCKY BLUEGRASS, AND THEIR HYBRIDS (*POA* SPP.)

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#### ABSTRACT

The fall armyworm Spodoptera frugiperda (J. E. Smith) (Lepidoptera: Noctuidae), is a destructive pest of many species of both C3 and C4 (cool- and warm-season) turfgrass. No-choice experiments were conducted to evaluate 13 turfgrass genotypes of various Poa spp. for susceptibility or resistance to the fall armyworm. All 13 genotypes, including 8 Texas bluegrass (Poa arachnifera Torr.), 2 Kentucky bluegrass (P. pratensis L.) and 3 Kentucky bluegrass × Texas bluegrass interspecies hybrids, were antibiotic and produced an accumulated >80% mortality of neonate larvae before they pupated. 'Reveille' (a hybrid) provided 100% antibiosis of larvae within 4 d of feeding. When 4-d-old fall armyworm larvae that had first fed on a susceptible Poa host were confined on the same 13 Poa genotypes as in the neonate test, a much higher survival rate was recorded. 'Reveille' produced 94.4% mortality after 3 d of feeding and 100% mortality after 8 d of feeding, while TXKY90-13-16 (another hybrid) provided 100% mortality of larvae within 13 d of feeding. A third hybrid, TXKY90-13-8 was one of the more susceptible genotypes. For the 4-d-old larvae, 'Baron' and 'Delwood Fine' Kentucky bluegrass provided only 50 and 22.2%, respectively, mortality after 8 d of larval feeding and did not produce 100% mortality until pupation. Also, mortality of larvae on the 8 Texas bluegrass genotypes produced ≤45% maximum accumulated mortality by pupation or adult emergence. 'Laser' rough bluegrass (*P. trivialis* L.) is an excellent host with  $\leq$  5.6% larval mortality.

Key Words: turfgrass pests, host plant resistance, antibiosis, *Poa arachnifera, Poa pratensis, Poa* hybrids

#### RESUMEN

El gusano cogollero Spodoptera frugiperda (J. E. Smith) (Lepidoptera: Noctuidae), es una plaga destructiva para muchas especies de céspedes tipo C3 y C4 (climas fríos y templados). Se llevaron a cabo experimentos de no-opción para evaluar la susceptibilidad o resistencia de 13 genotipos de varias especies Poa spp. al gusano cogollero. Todos los 13 genotipos, incluyendo 8 Texas bluegrass (Poa arachnifera Torr.), 2 Kentucky bluegrass (P. pratensis L.) y 3 híbridos interespecíficos de Kentucky bluegrass × Texas bluegrass fueron antibióticos y produjeron mortalidades >80% en larvas neonatas antes de que puparan. 'Reveille' (un híbrido) proveyó una antibiosis de 100% en los primeros 4 días. Cuando larvas de 4-días fueron primero alimentadas con hospederos susceptibles de Poa, y después confinadas a en los mismos 13 genotipos de Poa, se observó una mayor tasa de sobrevivencia. 'Reveille' produjo una mortalidad de 94.4% después de 3 días de alimentación y una mortalidad de 100% de después de 8 días, mientras que TXKY90-13-16 (otro híbrido) proveyó una mortalidad de 100% dentro de 13 días. Un tercer híbrido, TXKY90-13-8, fue el más susceptible de los genotipos. Para larvas de 4 días de edad, los cultivares de Kentucky bluegrass 'Baron' y 'Delwood Fine' proveyeron solo un 50 y 22.2% de mortalidad, respectivamente despu'es de 8 días de alimentación, y no produjeron mortalidades de 100% hasta pupación. Por otro lado, la mortalidad de larvas en los ocho genotipos de Texas bluegrass fue aproximadamente un 45% de la mortalidad máxima tanto en pupación o emergencia de los adultos. El césped 'Laser' rough bluegrass (P. trivialis L.) es un hospedero excelente, mostrando solo una mortalidad de 5.6%.

Translation provided by the authors.

Genetic plant resistance to pests including insects, mites, and diseases is an effective and economical control strategy and should be a major component of every Integrated Pest Management program when resistant cultivars are available. A major need in the turfgrass industry is the continued development of improved cultivars with inherent resistance to the primary turfgrass pests regardless of their utilization from production fields to installed landscapes, golf courses, or recreational fields. When pest resistant cultivars are used in the landscape, they will help to reduce the need for pesticide input into urban and suburban landscapes and indirectly reduce the potential for environmental contamination.

The fall armyworm (FAW) Spodoptera frugiperda (J. E. Smith) (Lepidoptera: Noctuidae) is a destructive pest of over 50 species of plants (Luginbill 1928) and it is known to feed on many species of both C3 and C4 (cool- and warm-season) turfgrass (Reinert et al. 1997, 1999). Leuck et al. (1968) first identified resistance to fall armyworm in bermudagrass. Lynch et al. (1983), Quisenberry & Wilson (1985), and Jamjanyu & Quisenberry (1988) confirmed the high level of antibiosis and nonpreference in Tifton 292 and several other bermudagrass genotypes. Wiseman et al. (1982) and Chang et al. (1985) reported a high level of resistance to fall armyworm in 'Common' centipedegrass Eremochloa ophiuroides (Munro.) Hack. and Reinert & Engelke (unpublished manuscript) discovered high levels of antibiosis in 'Cavalier' (Zoysia matrella L.) and several other genotypes of zoysiagrass. Cavalier was introduced as a new cultivar for its resistance to environmental stresses including its resistance to the FAW, other chewing insects, and several diseases. Resistance to FAW has been characterized among 46 Kentucky bluegrass (Poa pratensis L.) cultivars (Reinert et al. 2004b). The potential for use of insects and mites resistant turfgrass cultivars was summarized by Reinert et al. (2004a), but they also emphasize the considerable lack of known information on host response to insects and mites in the turfgrass ecosystem.

There is a need across the southern United Stated for a perennial cool-season grass for use as turf and for a permanent winter pasture for livestock. The existing cool-season perennial grasses (C3 grasses), primarily tall fescue (*Festuca arundinacea* Schreb.) that are available require higher rainfall and more temperate climates than the C4 grasses that are typically utilized throughout the southern United States. Texas bluegrass (*P. arachnifera* Torr.) is a C3 grass and its hybrids with Kentucky bluegrass (*P. pratensis* L.) have considerable potential in these drier and hotter regions (Read 1994, 2001).

The purpose of this research was to evaluate genotypes of Texas bluegrass, Kentucky bluegrass, and their interspecies hybrids and to identify potential resistance to the FAW, which is one of the primary pests of turfgrasses and forage grasses across the southern region and much of the Eastern and Midwestern U.S.

## MATERIALS AND METHODS

*Poa* genotypes and interspecies hybrids (Tables 1 and 2) were maintained in the greenhouse and cultured in plastic pots (15.24 cm. top diam., 12.7 cm bottom diam. by 17.7 cm tall) and fertilized bi-weekly with Peter's 20-20-20 (NPK) at approximately 170 ppm. Leaf and stem clippings from these plants were used to bioassay FAW larvae in no-choice laboratory feeding experiments.

Clippings were taken from several plants for each genotype and mixed together so that a representative sample of the grass was always used to feed the FAW larvae. Each experiment was set up in the laboratory with plastic Petri dishes (9-cm diam.  $\times$  20 mm deep) as feeding chambers for larvae. Each feeding chamber was provided with two water saturated 7.5-cm filter paper discs. Water was added to the filter paper as needed throughout the experiments to keep it saturated and maintain the grass cuttings. Each dish was provided with a small amount of fresh leaf tissue (ca. 3 g) of the respective *Poa* genotype.

Grass was added or replaced daily or everyother-day throughout the experiment so that turgid fresh grass was always available to the developing larvae. For these experiments, eggs of the corn strain of FAW were obtained from the lab colony maintained at the USDA-ARS-IBPMRL at Tifton, GA. Larvae were introduced into the feeding chambers as neonates within a few hours after hatching in Experiment 1. In Experiment 2, they were introduced as 4-d-old larvae that had been developed on fresh tissue of 'Laser' rough bluegrass (*Poa trivialis* L.). This grass serves as an excellent host, usually with near 100% survival.

For the first experiment, 3 neonate larvae were randomly selected after egg hatch and placed on each grass in the feeding chambers in each replicate (Table 1), and dishes were arranged in a randomized complete block design with 6 replicates on the laboratory bench. Since FAW egg masses are usually laid on some structure or debris adjacent to the turf area and the larvae then migrate to the turf setting to feed, we also evaluated 4-dold larvae on each of the test genotypes. For the second experiment, neonate larvae were allowed to develop for 4 d on leaf clipping of Laser rough bluegrass. When larvae were 4-d-old, 3 larvae were randomly selected and placed in the feeding chambers with the respective Poa genotypes (Table 2) in a randomized complete block design with 6 replicates.

For both experiments, survivorship was recorded when clippings were added either daily or every-other-day until pupation and at adult emergence. All surviving larvae were weighed when 12-d-old, well before any pupation occurred and all pupae were weighed within 24 h after pupation. Days from egg hatch to pupation and adult emergence were recorded.

#### Statistical Analysis

Data were analyzed by analysis of variance procedures (ANOVA and GLM) and means separated by Tukey's studentized range (HDS) test (P = 0.05) (SAS Institute 2008). Mortality data were transformed to arcsine (x + 0.001) before each ANOVA was performed, but the actual percentage for mortality is presented.

Cultivar or Genotype	7-d-old Mortª	$\begin{array}{l} 12\text{-d-old} \\ wt \ (mg)^{\scriptscriptstyle b} \end{array}$	12-d-old % mort ª	Pupa wt (mg)°	Days to pupa <sup>d</sup>	Pupa % mortª	Adult days <sup>e</sup>	Adult % mort ª
Kentucky bluegrass ( <i>Poa pratensis</i> ) Delwood Fine	100 a <sup>f</sup>	g	100 a	_		100 a		100 a
Baron	72.2 a	$22.8 a^{\rm f}$	72.2 a	_	_	100 a	_	100 a
TX x KY hybrids (Poa pratensis x Poa arachnifera)								
Reveille	100 a		100 a	_	_	100 a	_	100 a
TXKY90-13-16	100 a		100 a	—	_	100 a		100 a
TXKY90-13-8	83.3 a	66.9 ab	83.3 a	237.1 a	23.3 b	83.3 a	$37.3 \mathrm{b}$	83.3a
Texas bluegrass (Poa arachnifera)								
TBPC15-10	100 a		100 a	_	_	100 a		100 a
TBPC20-16	100 a	_	100 a	_	_	100 a	_	100 a
TBPC27-7	100 a	_	100 a	_	_	100 a	_	100 a
Syn3	100 a	_	100 a	_	_	100 a	_	100 a
Syn4	100 a	_	100 a	_	_	100 a	_	100 a
Syn5	94.4 a	3.2 a	94.4 a	184.6 ab	35.0 a	94.4 a	47.0 a	94.4 a
Tejas 1	94.4 a	6.0 a	94.4 a	208.4 ab	26.0 b	94.4 a	31.0 c	94.4 a
Syn2	83.3 a	28.7 a	83.3 a	149.7 b	$25.0 \mathrm{b}$	88.9 a	30.0 c	94.4 a
Rough bluegrass (Poa trivialis)								
Laser	5.6 b	149.0 b	5.6 b	212.9 ab	19.5 c	5.6 b	33.6 bc	5.6 b

<sup>a</sup>Mean % accumulated mortality at 7 d old, 12 d old, pupation, and at adult emergence on each grass genotype.

<sup>b</sup>Mean weight of 12-d-old larvae after 8 d of feeding.

<sup>c</sup> Mean pupa weight taken within 1 d of pupation. <sup>d</sup> Mean number of days from egg hatch to pupation.

<sup>e</sup> Mean number of days from egg hatch to adult emergence.

'Means in a column followed by the same letter are not significantly different by

Tukey's Studentized Range (HSD) test (P = 0.05)

<sup>s</sup>No larvae survived to this growth stage.

TABLE 2. RESISTANCE IN BLUEGRASSES TO	FEEDING BY 4-DAY-OLD FALL ARMYWORM	LARVAE (MEANS OF 6 REPLICATES)
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Cultivar or Genotype	7-d-old Mortª	$\begin{array}{c} 12\text{-d-old} \\ wt \ (mg)^{\flat} \end{array}$	12-d-old % mort <sup>a</sup>	Pupa wt (mg)°	Days to pupa <sup>d</sup>	Pupa % mort <sup>a</sup>	Adult days <sup>°</sup>	Adult % mortª
Kentucky bluegrass (Poa pratensis)								
Delwood Fine	$16.7 \ bc^{ \rm f}$	22.1 a <sup>s</sup>	22.2 c	g		100 a	_	100 a
Baron	32.2  bc	25.4 a	$50.0 \mathrm{b}$	—	—	100 a	—	100 a
TX x KY hybrids (Poa pratensis x Poa arachnifera)								
Reveille	94.4 a	_	100 a	_	_	100 a	_	100 a
TXKY90-13-16	44.4 b	5.6 a	83.3 ab	_	_	100 a	_	100 a
TXKY90-13-8	11.1 bc	66.6 b	11.1 c	229.1 c	24.4 d	16.7 bcd	38.1 d	16.7 bcd
Texas bluegrass (Poa arachnifera)								
TBPC15-10	$11.1 \mathrm{\ bc}$	14.5 a	11.1 c	187.4 a	33.8 a	27.8 bcd	48.3 a	27.8 bcd
TBPC20-16	5.6  bc	20.1 a	5.6 c	193.1 a	29.3 с	16.7 bcd	43.2 c	16.7 bcd
TBPC27-7	5.6  bc	17.1 a	5.6 c	182.9 a	30.8 bc	$5.6  ext{ cd}$	45.0  bc	5.6 cd
Syn3	11.1  bc	18.5 a	11.1 c	181.3 a	30.9 bc-	44.4 b	45.6 abc	44.4 b
Syn4	$5.6 \ \mathrm{bc}$	16.4 a	16.7 c	172.6 a	33.6 а	33.3 bcd	27.6 ab	33.3 bcd
Syn5	$5.6 \ \mathrm{bc}$	15.6 a	5.6 c	179.9 a	32.1 ab	33.3 bcd	46.3 ab	33.3 bcd
Tejas 1	0 c	18.4 a	5.6 c	$201.7 \mathrm{b}$	29.1 с	$5.6  ext{ cd}$	43.2 c	5.6 cd
Syn2	$5.9 \ \mathrm{bc}$	16.8 a	17.8 c	197.1 ab	31.5 abc	38.9 bc	45.6 abc	38.9 bc
Rough bluegrass (Poa trivialis)								
Laser	0 c	139.4 c	0 c	229.9 с	21.9 e	0 d	35.7 d	0 d

"Mean % accumulated mortality at 7 d old, 12 d old, pupation, and at adult emergence on each grass genotype.

<sup>b</sup>Mean weight of 12-d-old larvae after 8 d of feeding.

<sup>e</sup>Mean pupa weight taken within 1 d of pupation.

<sup>d</sup>Mean number of days from egg hatch to pupation.

<sup>e</sup>Mean number of days from egg hatch to adult emergence.

Means in a column followed by the same letter are not significantly different by Tukey's Studentized Range (HSD) test (P = 0.05)

<sup>s</sup>No larvae survived to this growth stage.

## RESULTS

## Neonate Larvae

Each Poa genotypes, except P. trivialis, was highly antibiotic to the neonate FAW larvae. All Texas bluegrass, Kentucky bluegrass, and Texas bluegrass × Kentucky bluegrass hybrids produced >61% mortality within 4 d of feeding (Table 1). Five of the grasses, 'Reveille' (TXKY90-16-1) a bluegrass hybrid, Syn4, TBPC15-10 and TXPC27-7 Texas bluegrasses, and 'Delwood Fine' Kentucky bluegrass each provided 100% mortality within 4 d. After 7 d of feeding, TXPC20-16 and Syn3 provided 100% mortality, and after 12 d, TXKY90-13-16 also provided 100% mortality of the confined larvae. 'Baron' Kentucky bluegrass did not produce 100% mortality until after larvae had fed for 17 d. All of the grasses except Laser rough bluegrass provided >80% and statistically significant mortality before larvae were able to pupate. Laser was an excellent host for the FAW with only 5.6% mortality of the larvae started as neonates, which may be within the expected mortality in nature. Differences in larval weights at 12 d were also significantly different (Table 1). Larvae that fed on Laser were much heavier than those that fed on any of the other *Poa* genotypes. The larvae that did survive and mature to adults on TXKY90-13-8 bluegrass hybrid, however, were only about one half the sizes of those that developed on Laser (Table 1). Larvae that pupated after developing on TXKY90-13-8 were the largest and produced significantly heavier pupae than those that developed on Syn2. The larvae feeding on Laser pupated in the shortest feeding period (19.5 d) while those feeding on the other Poa genotypes took 4.0 to 15.5 d longer. Individuals that successfully emerged as adults on Syn5 took 47 d in contrast to only 33.6 d for the larvae that developed on Laser. Even though 1 to 3 individuals per genotype were able to pupate and reach adult emergence on TXKY90-13-8, Tejas1, Syn2 or Syn5, these grasses should be considered as poor hosts for neonate larvae of FAW because of the sublethal antibiosis exhibited.

## 4-day-old Larvae

The survival of larvae was much higher if they first fed for 4 d on the very palatable host, Laser before being confined in the no-choice feeding study with each of the 14 *Poa* genotypes (Table 2). 'Reveille' hybrid bluegrass produced highly significant antibiosis (94%) of the larvae after only 3 d of feeding (7-d-old) and all larvae were dead within 12 d. Another hybrid, TXKY90-13-16, provided 100% mortality after 13 d of feeding (17-dold), whereas, the third hybrid, TXKY90-13-8 was one of the most susceptible *Poa* genotypes evaluated and only caused 11.1% mortality of larvae after 13 d of feeding and only 16.7% mortality by pupation and adult emergence. The 2 cultivars of Kentucky bluegrass, Baron and Delwood Fine, provided only 50 and 22.2%, respectively, mortality after 8 d of feeding (12-d-old) and neither produced 100% mortality until pupation. None of the *P. arachnifera* genotypes produced <25% mortality of larvae and <45% maximum mortality at pupation or adult emergence (Table 2). The cultivar 'Tejas1' and 2 other Texas bluegrass genotypes (TBPC27-7, TBPC20-16) and 1 hybrid (TXKY90-13-8) showed no resistance (<17% mortality) and should be considered highly susceptible to the FAW. No mortality was recorded on Laser rough bluegrass during this study.

Similar to the results in the previous experiment that started with neonate larvae, the larvae in this study developing on TXKY90-13-8 were also ca. 3 times larger than those developing on the other Poa genotypes, except for those on the highly susceptible Laser rough bluegrass which were about 7 times larger (Table 2). Additionally, the 12-d-old larvae feeding on Laser in both studies were about the same weight and required similar time periods to reach adult emergence. Even though survivorship was relatively high on most of the *Poa* genotypes, the significantly reduced larval and pupal weights are an indication of sublethal (antibiotic) resistance. Individuals confined on most of these genotypes also required 7 d or longer before pupation and adult emergence compared to those developing on Laser which required an average of only 21.9 d to pupation and only 35.7 d before adult emergence.

Another mechanism of resistance was observed in larvae feeding on 'Baron' and 'Delwood Fine' Kentucky bluegrass and on 'Reveille' and TXKY90-13-16 hybrids. Larvae feeding on these genotypes would develop normally to either the second or third instar, when they would begin swelling during ecdysis and appear to freeze in this bloated state and they were unable to complete the molt or the shedding of the old larval skin. Since this phenomenon was only observed with larvae feeding on genotypes of Kentucky bluegrass and hybrids between Kentucky bluegrass and Texas bluegrass, it is assumed that this mechanism of resistance is inherited from the Kentucky bluegrass parents and was transferred to 'Reveille' and TXKY90-13-16 during hybridization.

#### DISCUSSION

Results of this study indicate that a strong level of resistance to the FAW is present in Kentucky bluegrass cultivars, Baron and Delwood Fine. The resistance level to FAW, however, can vary among Kentucky bluegrass cultivars. Wabash, Adelphi, Eagleton, and Monopoly each produced >90% mortality after only 7 d of feeding, while Kenblue, PTDF22B2, and Glade did not produce grater than 30% mortality even at adult emergence (Reinert 2004b). The level of susceptibility varied considerably among the Texas bluegrass genotypes with Syn3 providing the higher level of resistance (44.4% mortality) to the 4-d-old larvae. Two of the Texas bluegrass  $\times$  Kentucky bluegrass hybrids ('Reveille' and TXKY90-13-16) inherited the high level of resistant (100% mortality) while the third hybrid, TXKY90-13-8, is susceptible. Laser rough bluegrass is highly susceptible to FAW and should be used as a standard for comparison in other studies with turfgrasses to document their levels of susceptibility or resistance. Additional feeding studies with FAW larvae on Kentucky bluegrass are needed to characterize the true nature of the ecdysis mechanism of resistance.

There appears to be 2 different mechanisms for antibiosis in the bluegrasses. One mechanism was observed in Baron and Delwood Fine Kentucky bluegrass and in the 2 Texas bluegrass  $\times$ Kentucky bluegrass hybrids, Reveille and TXKY90-13-16. In this case during the second or third instar, the larvae begin to swell during ecdysis and appear to freeze in the bloated state thus causing death. This antibiosis would most likely be due to some toxin associated with Kentucky bluegrass. The other antibiosis mechanism observed in Texas bluegrass and the hybrid TXKY90-13-8 may to be related to poor quality diet. In this case, the antibiosis appears related to poor digestibility or palatability of the plant material as opposed to some toxic compound. Further studies with FAW resistance will address this hypothesis.

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