Screening and Breeding for Bermudagrass Stem Maggot (BSM) Resistance Using U.S. Bermudagrass Germplasm

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Key words: Bermudagrass Stem Maggot; tolerance; breeding

Abstract
Bermudagrass (*Cynodon* sp.) is an important perennial forage grass grown in many parts of the world. Bermudagrass Stem Maggot (BSM) (*Atherigona reversura* Villeneuve) is an insect pest that reduces forage yield and nutritive value if it is not controlled. The pest, native to SE Asia, was first documented in North America in 2009 and is now considered invasive. A collection of over 300 forage bermudagrass accessions was evaluated in the field for susceptibility to BSM in 2014 and 2015. Tolerant lines and susceptible checks were then evaluated for yield loss due to BSM in a replicated field study by comparing insecticide-sprayed plots to unsprayed plots in Tifton, GA starting in 2016 continuing through the summer of 2019. For mid to late summer harvests during 2017, BSM reduced yield of Alicia and Russell by over 40% and Tifton 85 by up to 35%. However, tolerant accessions exhibited less than 10% yield loss and had dry matter yields comparable to Tifton 85. Nutritive value will also be assessed. These accessions will be further evaluated and used in plant breeding.

Introduction
Bermudagrass (*Cynodon dactylon*) is a primary forage crop for the Southeastern US. It is a perennial warm-season forage grass that covers millions of hectares of pasture for grazing and hay production. However, it has been invaded by the invasive bermudagrass stem maggot (BSM) has damaged bermudagrass pastures and hayfields throughout the Southeast (Baxter et al., 2014). The adult is a small fly that can travel to neighbouring pastures and lay eggs on the bermudagrass leaf. Once hatched, larvae feed outwards from the terminal node of the plant and kill the top 2-3 leaves on the stem, stopping growth of the damaged tillers and reducing the number of tillers on the plant (Baxter et al., 2014). Since 2010, BSM has spread throughout the South-eastern US, damaging bermudagrass hayfields and pastures as far north as North Carolina and Kentucky and as far west as Texas (Baxter et al., 2014). BSM has also been observed on stargrass in Central America in recent years (personal observation). Stargrass (*Cynodon nlemfuensi* Vanderyst) and cultivars such as 'Tifton 68' [Burton and Monson, 1984] or 'Tifton 85' [Burton et al., 1993] were shown to have less BSM damage than fine-textured bermudagrass (*Cynodon dactylon* (L.) Pers.) varieties such as 'Coastal', 'Alicia', and 'Russell' (Baxter et al., 2015). Additional field work verified that fine-stemmed bermudagrass lines are more susceptible and that Alicia, Coastal and Russell had yield losses as high as 60% due to BSM during late summer harvests (Baxter et al. 2019). Tifton 85 suffered enough yield loss to warrant the need for new cultivars that are more resistant to or tolerant of BSM. Thus, a screening of available bermudagrass germplasm is necessary for breeding and development of improved cultivars. This study used the bermudagrass core collection (Anderson, 2005) and additional germplasm to screen for BSM tolerance.

Methods and Study Site
A collection of over 300 forage bermudagrass accessions was evaluated in the field for susceptibility to BSM among other traits. The 1 m^2 plots were established using vegetative material that originated from a core collection from Tifton, Ga (Anderson, 2005) and additional germplasm from USDA-GRIN. Pots (10 cm) were established from a single sprig during the fall of 2014 and transplanted into two randomized replications at three locations (Tifton, Ga; Citra, FL; Ona, FL) in the spring of 2015. Visual ratings (0 = no visible damage to 5 = over 80% damage to upper 3 leaves) were made for two years (2015 and 2016) at Citra and Tifton and during 2015 at Ona. Plots were harvested 5 times in 2015 and four times in 2016 at Tifton, GA. The 15 most tolerant (rating < 2) with the best yield were advanced for further analysis. Pots (10 cm) were established using vegetative material of the 15 lines plus 5 checks (Tifton 85, Alicia, Russell, Jiggs and stargrass) in the spring

of 2016 and transplanted to plots (2 m x 6 m). Plots were arranged in four pair-plot randomized replications. Entries were evaluated for yield loss due to BSM by comparing sprayed to unsprayed plots within the paired plots in Tifton, GA starting in 2016, through the summer of 2019. Plots were harvested once in the fall of 2016, 5 times in 2017 and 2018, and four times in 2019. Yield of each plot was recorded, and an approximately 300 g sample retained from each plot. Samples were weighed wet, dried at 50º C, and weighed dry to determine dry matter content. Samples were then ground with a Wiley Mill, then a Cyclone mill and evaluated by Near Infrared Spectroscopy (NIRS) with a FOSS 6400 spectrometer from a calibration for in vitro dry matter digestibility (IVDMD), neutral detergent fibre (NDF) and acid detergent fibre (ADF). NIR equation accuracy at this lab has been verified by the National Forage Testing Association (NFTA, 2019) and all prediction models were provided by the NIRS Consortium (NIRSC, 2019). Data were analysed using Proc Mixed in SAS 9.4 (Littell, 2006).

Results

Reduction in Yield from BSM

Susceptible cultivars experienced a large yield loss primarily in hay harvests from July through October. Some of the greatest losses were observed during September (Table 1). A number of accessions had significantly less yield loss than current cultivars. Previously, Tifton 85 was identified as one of the most tolerant cultivars (Baxter et al. 2019). However, many PI accessions were more tolerant than Tifton 85 to BSM damage.

Table 1: The percent reduction in yield of bermudagrass plant introduction (PI) and cultivars due to BSM in September of years 2016 to 2108 in Tifton, GA, USA (Same letters in a column indicate non- significant differences at p = 0.05 level).

Entry	2016 % Red.	2017 % Red.	2018 % Red.	Mean
PI 294467	6.6 a	**17.0 a-c**	14.2 a	**12.5**
PI 290901	7.7 ab	**19.1 a-d**	**5.6 a**	**10.8**
PI 290812	9.5 ab	**18.8 a-d**	26.6 a-e	**18.3**
Breeding Line	10.3 ab	**15.1 ab**	**9.6 a**	**11.7**
PI 290664	11.1 ab	**12.3 a**	19.5 abc	**14.3**
PI 290872	19.2 ab	**16.2 a-c**	**8.0 a**	**14.5**
Tifton 85	18.5 ab	51.1 fg	24.4 a-d	31.3
Jiggs	47.4 c	57.4 gh	31.1 b-f	45.3
Alicia	40.3 c	70.1 h	59.4 f	56.6

Yield Potential of BSM tolerant accessions

The most tolerant accessions also produced the most hay biomass over years at Tifton, GA (Table 2). These accessions have generally coarse stems but less coarse than Tifton 85.

Table 2: Total dry matter yield (kg/ha) of bermudagrass plant introduction (PI) and cultivars in unsprayed plots from 2017-2019 at Tifton, GA. (Bold numbers in a column indicate significant differences from Tifton 85 at p = 0.05 level)

Entry	2017	2018	2019	Mean
PI 290664	**21112**	**26136**	**27914**	**25054**
Breeding Line	**20201**	**26224**	26946	**24457**
PI 290901	**19523**	**25024**	26167	**23571**
PI 294467	**19748**	**25428**	24364	**23180**
PI 290812	17987	22681	22326	20998
Tifton 85	16318	20534	25066	20639
Jiggs	13399	16955	19111	16488
Russell	9987	13694	19877	14519
Alicia	8791	14250	16509	13183
Mean	13033	17140	18467	16213

MSD	1781	2386	2658	2025

Discussion

Though complete resistance to BSM has not been found, several bermudagrass plant introductions were identified with significantly more tolerance then current cultivars and will be used for direct release and for breeding. It is important to note that four accessions had significantly higher yields than Tifton 85 in 2017 and 2018 under no spray conditions. These accessions will be further evaluated for forage nutritive value.

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