Release of a New Forage Bermudagrass Cultivar from the USDA-NPGS Cynodon Collection

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RELEASE OF A NEW FORAGE BERMUDAGRASS CULTIVAR FROM THE USDA-NPGS CYNODON COLLECTION

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Key words: Herbage accumulation; Nutritive value; Bermudagrass Stem Maggot; Tifton 85; Newell

Abstract

Warm-season perennial grasses are the backbone of the pasture-based livestock industry in the southeastern USA. In Florida specifically, bahiagrass (*Paspalum notatum* Flugge) and bermudagrass (*Cynodon* spp.) support 1 million head of cattle and 15,000 beef cattle operations. Bermudagrass is the most widely planted forage species in the southeastern USA, planted in approximately 15 million ha and used for grazing, hay and silage. The genus *Cynodon* is native to southern Africa and germplasm collections have revealed a high degree of genetic variability within the genus. The United States Department of Agriculture National Plant Germplasm System (USDA-NPGS) maintains a collection of bermudagrass plant introduction (PIs) in Griffin, GA, USA and the USDA Georgia Coastal Plains Experiment Station, Tifton, GA, maintains additional forage germplasm. Multi-location trials were established in 2014 in four states (FL, GA, NC and OK) to screen the collection for herbage accumulation (HA) and nutritive value (NV). Due to the large genotype by environment interaction for HA across states, we focused on selecting accessions adapted to South Georgia and Florida. Several PIs showed improved HA and NV compared to 'Tifton 85' across several trials and years. Particularly, PI 316510 produced high HA in Citra, FL and Tifton, GA, had improved NV traits, and faster establishment compared to Tifton 85. We confirmed that PI 316510 is tetraploid by chromosome counts and flow cytometry. The PI 316510 has been released by the University of Florida under the name "Newell".

Introduction

Bermudagrass (*Cynodon dactylon* L.) is the most widely planted forage species in the southeastern USA, covering approximately 15 million ha (Taliaferro et al. 2004). The species is used primarily as a forage for grazing, hay or silage, and it is also of major importance as a turfgrass for home lawns, sports fields, and on golf course fairways and tees. Bermudagrass is preferred over other warm-season perennial forages because of its greater HA, NV, and persistence under diverse climatic conditions and management practices. *Cynodon dactylon* is the most prevalent species, and together with *C. nlefuensis* Vanderyst (stargrass) are the two most important species for forage production (Sollenberger et al. 2008). Stargrass is a robust non-rhyzomatous type, while bermudagrass is rhizomatous, and these two species are cross-compatible (Sollenberger et al. 2008). There have been extensive efforts placed in breeding bermudagrass that resulted in multiple cultivar releases (Burton et al. 1967, 1972, 1984, 1993). Interspecific hybrids between bermudagrass and stargrass have provided vital ecosystem services for decades and revolutionized the livestock and hay industry in the region (Corriher and Redmon 2009). In Florida, stargrass cultivars have been utilized, including 'Ona', 'Florona', and 'Florico', and 'Mislevy' is a new bermudagrass cultivar released in 2019 (Vendramini et al. 2020).

The growing needs for animal-based products in a world facing competing demands, climate variability, and emerging pests, require investments in research on forage germplasm enhancement to breed novel cultivars that will provide an array of ecosystem services. Bermudagrass stem maggot (BSM) (*Atherigona reversura* Villeneuve) was first discovered in the United States in 2009 in Los Angeles, CA, and it has since spread throughout the southeastern United States and areas worldwide (Ribeiro et al. 2016; Patitucci et al. 2016; Baxter et al. 2019). The BSM damages all bermudagrass cultivars, leading to yield reductions of up to 50 percent (Baxter et al. 2019).

In the southeastern USA, warm-season perennial grass productivity is limited during early spring and late autumn due to, among several factors, the most critical being lower temperature (Ball et al. 1996). Hence, there is a need to develop cultivars with increased production during these periods to increase farmer's profitability by decreasing the needs for stored forage and/or feeding supplementation and extend the grazing season and hay harvest season. Therefore, development and release of a bermudagrass cultivar with superior HA and NV, and BSM tolerance would provide economic benefits to livestock and forage producers.

Methods and Study Site

Study 1. Germplasm screening in multiple states in the southeastern USA

A collection of 286 accessions of *Cynodon* spp. were planted in replicated trials in Citra, Ona, and Marianna, FL, Tifton, GA, Jackson Springs, NC, and Ardmore, OK (Fig.1 A). Plots were harvested to determine HA and number of harvests ranged from three (Jackson Springs) to seven (Citra) per year. Additionally, four NV traits were measured in Citra, FL using wet chemistry analysis for 11 harvests for a selected group of 15 genotypes (Lopes de Souza et al. 2020). Visual ratings for BSM damage were collected in Ona and Citra, FL and Tifton, GA (Fig.1 B). Variance components were estimated using linear mixed models and genetic parameters are reported for HA. Significant effects of variance components were tested using a Likelihood Ratio Test (LRT).

Study 2. Additional small-plot experiments in Florida

2.1 Multi-location trials were planted in spring 2017 in Ona, Marianna, and Hague, FL. Plugs were propagated for each genotype and used for establishing the experiments under a RCBD with 10 genotypes and 4 blocks (1.8 x 4.6 m plot size). Herbage accumulation was measured five times in each location in 2018 and 2019.

2.2 Establishment trial planted in Gainesville, FL on 5/28/2020 using Tifton 85, Jiggs, Mislevy, PI 316510 and Entry 286. Plots (1.5 x 12 m) were established using approximately 1600 kg ha^{-1} of fresh cut tops (>6 weeks growth), following a RCBD with four replicates and a split-plot treatment arrangement. Herbicide treatments: dicamba (279 g active ingredient [a.i.] ha^{-1}) + 2,4-D (799 g a.i. ha^{-1}; Weedmaster®, 7 days after planting - DAP), aminopyralid (92.7 g a.i. ha^{-1}) + florpyrauxifen-benzyl (9.3 g a.i. ha^{-1}; Duracor®, 7 DAP), sulfosulfuron (52.5 g a.i. ha^{-1}, Outrider®, 14 DAP), and a control. Plots were fertilized with 37, 9 and 28 kg ha^{-1} of N, P_2O_5 and K_2O 30 DAP, and 60, 16 and 47 kg ha^{-1} of N, P_2O_5 and K_2O 60 days after planting. Plots were visually assessed 30, 60 and 90 DAP for bermudagrass cover using a 0-5 scale, where 5 equal 100% cover.

Study 3. Ploidy determination

Flow cytometry was used to evaluate genome size of PI 316510 and other bermudagrass lines were used as internal standards. Chromosome counting in root tips were conducted in PI 316510 to confirm its ploidy.

Results

Study 1. Germplasm screening in multiple states in the southeastern USA

The genetic variance for HA was significant (LRT $P<0.05$) in all locations for the single-location analysis, except for Marianna, FL; thus, Marianna was excluded from the multi-location analysis. The broad-sense heritability (H^2) estimate for HA for the multi-location model was 0.12±0.02, and the genotype by location correlation was 0.32. These two parameters indicated the presence of genetic variation in the whole collection and showed a strong genotype by environment interaction for HA. A principal component analysis (PCA) analysis performed with genotypic values estimated for HA across four locations (Citra, FL, Tifton, GA, Jackson Springs, NC, and Ardmore, OK) demonstrated a clear genotype by environment effect for HA (Fig. 1 C). Some genotypes produced higher HA in Florida (entry 322: PI 316510), compared to genotypes more adapted to higher latitudes (entries 283, 237, 256). Tifton85, produced high HA across most locations.

Figure 1. (A) Replicated germplasm screening for herbage accumulation (HA) using 286 accessions of *Cynodon* spp. across the southeastern USA. (B) Bermudagrass stem maggot is a new threat to bermudagrass pastures in the southeastern USA. (C) Principal component analysis (PCA) showing a large genotype by environment interaction for HA across four locations (GNV: Gainesville, FL; GA: Tifton, GA; OK: Ardmore, OK; NC: Jackson Springs, NC).

The HA based on dry matter rankings is presented in Table 1 for a select group of entries and controls, for the data collected in Citra, FL and Tifton, GA. In this comparison, PI 316510 is the highest yielding line at Citra in 2015 and 2016, and it produced higher HA than all controls in both years. At Tifton, GA the HA for PI 316510 were lower than Tifton85 in 2015, but similar in 2016 (data not presented).

Table 1. Predicted herbage accumulation (HA, kg/ha), crude protein (CP), phosphorous concentration (P), in vitro digestible organic matter (IVDOM), and neutral detergent fiber (NDF) for PI 316510, Florida 44 and Tifton 85 in Citra, FL.

Genotype	HA-2015	HA-2016	CP $g.kg^{-1}$	P $g.kg^{-1}$	IVDOM $g.kg^{-1}$	NDF $g.kg^{-1}$
PI 316510	21,450*	21,150	139 a	3.2 a	557 a	662 a
Florida 44	17,020	17,250	127 a	2.9 ab	461 b	667 a
Tifton 85	18,100	19,480	133 a	2.8 b	541 a	693 b
	Harvests = 5	Harvests = 6	Harvests =11	Harvests =11	Harvests =11	Harvests =11

**HA are predicted values across 286 accessions and post-hoc comparisons were not conducted. Nutritive value traits are means for 11 harvests and Tukey HSD Test was performed among 15 selected accessions. Means with same letter do not differ statistically (P≤0.05). For more details: Souza et al. 2020.*

All entries were visually rated for BSM and none of the entries exhibited complete BSM resistance. Tifton85 had the lowest BSM damage across commercial cultivars, while Coastal, FL44 and Jiggs ranked among the most susceptible (Figure 3). PI 316510 had similar BSM damage to Tifton 85 and Mislevy.

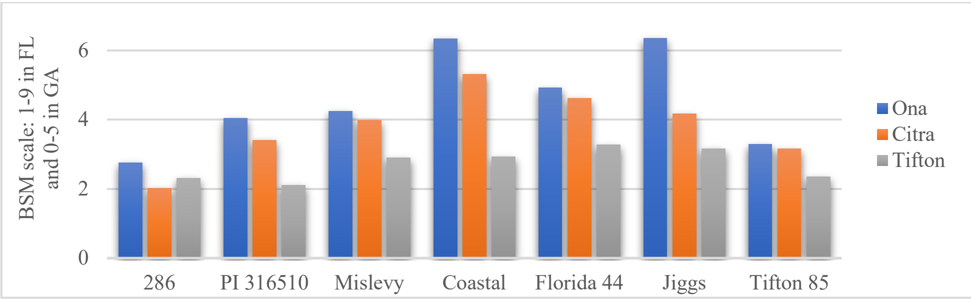


Figure 2. Bermudagrass stem maggot ratings across three locations. Rating scale of 1 to 9 (scale: 1 = no visible damage; 9 = more than 90% damage) and in Tifton on a scale of 0 to 5 (scale: 0 = no visible damage; 5 = more than 90% damage). These are predicted values estimated for 286 accessions and post-hoc comparisons were not conducted.

Study 2. Additional small-plot experiments in Florida

2.1 The averaged HA over 10 harvests (five in 2018 and five in 2019) is presented in Figure 3-A. PI 316510 was superior to Tifton 85 in total HA averaged over the two years in Hague and Marianna. Tifton 85 is currently among the most popular cultivar being planted on new fields in Florida and South Georgia. These data show considerable location variability for total seasonal yields.

Plots were assessed for bermudagrass cover 30, 60 and 90 DAP in Gainesville, FL (Figure 3-B). The PI 316510 has shown a much faster establishment rate than Tifton85 and other controls (Figure 3-B).

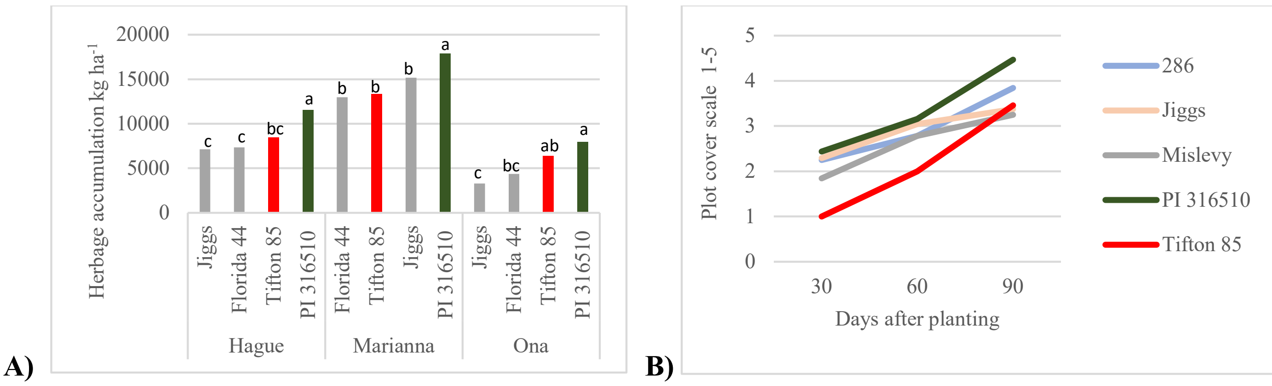
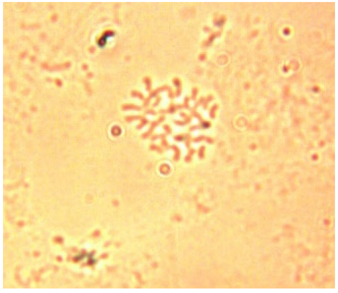


Figure 3. (A) Average herbage accumulation (HA; kg ha^{-1}) per year in Citra, Ona, and Marianna FL across 10 harvests performed between 2017 and 2018. (B)Bermudagrass plot cover 30, 60 and 90 days after planting in Gainesville, FL.

Study 3. Ploidy determination

It was determined that the ploidy of PI 316510 is 2n=4x=36 using chromosome counts (Figure 4-A), and by comparing its genome size with other known tetraploid bermudagrass cultivars (Figure 4-B).

A) 

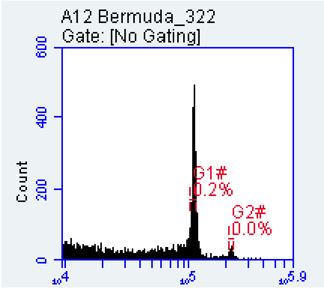
B) 

Figure 4. (A) Chromosomes of PI 316510 root tips under 400x magnification (2n = 4x = 36) and (B) histogram of PI 316510 depicting the propidium iodide florescence area signals (FL2A) of the sample nucleic DNA.

Discussion and Conclusions

The PI 316510 was obtained from the USDA-NPGS and was tested in multi-location/multi-year trials, showing higher total HA and improved NV, faster establishment using tops, and similar BSM tolerance compared to Tifton 85. The PI 316510 is tetraploid and could be used as a parent in crosses to continue the development of improved bermudagrass cultivars. The proposed cultivar name for PI 316510 is 'Newell', in honor of Dr. Wilmon Newell's legacy in Florida and the University of Florida.

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