Registration of 'Pristine' Zoysiagrass

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ABSTRACT

'Pristine' (Reg. No. CV-251, PI 652481) zoysiagrass [*Zoysia japonica* Stued. by *Zoysia tenuifolia* (L.) Merr.] was developed by the Florida Agricultural Experiment Station at the Everglades Research and Education Center, Institute of Food and Agricultural Sciences, University of Florida, Belle Glade, FL, and initially approved for release in 2005. This zoysiagrass variety originated as an open-pollinated progeny from 'Emerald' and tested in Florida under experimental designation BA-305. Pristine was selected for improved agronomic and horticultural traits, including reduced production of seed heads, finer leaf texture, darker leaf color, and a faster rate of ground coverage and crop establishment in southern Florida. In comparison to the standard variety Emerald, Pristine exhibited a 46% average annual reduction in seed-head production and generally produced seed heads with an attenuated morphology. It also produced darker green leaves that were 21% shorter and 19% narrower than Emerald, which visually conferred upon Pristine a more refined canopy structure and texture. In addition, ground coverage and crop establishment was significantly faster for Pristine at two of the three test sites. Pristine is primarily intended for use in the Florida specialty market for zoysiagrass.

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The zoysiagrasses (*Zoysia* spp.) are indigenous to the region around the western Pacific basin and one of a few underutilized warm season turf species grown in the southeastern United States (Beard, 1973). Currently,

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throughout Alabama, Florida, and Georgia there are 13 zoysiagrass varieties grown on 779 ha as either Foundation, Registered, or Certified seed (Southern Seed Certification Association, 2007; Georgia Crop Improvement Association, 2007). Zoysiagrass first gained wider acceptance in the United States with the release of 'Emerald', a standard "narrow-leafed" type zoysiagrass hybrid [Z. japonica Stued. × Z. tenuifolia (L.) Merr.] (Forbes et al., 1955), and its companion 'Meyer' zoysiagrass (Z. japonica Stued.), a "wideleafed" type (Anonymous, 1952). Meyer was developed and released by the USDA, while the Georgia Agricultural Experiment Station and the USDA jointly released Emerald. Both varieties exhibited adaptation along the U.S. eastern seaboard. Zoysiagrass has never achieved the anticipated level of market penetration, despite its utility for a number of unique turf applications (Beard, 1973). Nevertheless, for the last half-century the zoysiagrass industry has endured as a small but persistent and stable sector of the specialty turf market.

The purpose of this breeding program was to select and develop an alternative to the standard variety Emerald and to potentially enlarge the diversity of narrow-leafed zoysiagrass varieties grown in the southeastern United States. Pristine was selected for improved agronomic and horticultural traits, including reduced production of seed heads, a finer leaf texture as defined by shorter and narrower leaves, a darker leaf color, and a faster rate of establishment and ground coverage in southern Florida. It is a warm-season clonally propagated perennial variety intended for use in specialized turf applications such as golf course tee boxes, aprons around golf greens, croquet lawns and other exotic microhabitats.

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Methods

Pristine originated as an open-pollinated progeny derived from a version of Emerald, an interspecific hybrid between Z. japonica Stued. by Z. tenuifolia (L.) Merr. Pristine was first identified and selected in Palm Beach County, FL, and designated as breeding line BA-305. It was subsequently developed and evaluated in three phases. In the first phase, it was selected as a distinct individual from a group of individuals derived from Emerald. These open-pollinated individuals were originally selected to represent the variability and diversity of phenotypes in a planting of Emerald. From this initial group of individuals, visual selection was practiced for horticultural and agronomic traits including leaf color and texture along with seed-head production and the rate of ground coverage. In the second phase, a set of 15 individuals was selected, clonally propagated, and advanced for evaluation as unreplicated plots in Charlotte County, FL. In the third phase, a set of elite clones was evaluated under standard production practices in a three-replicate randomized block design at test sites in Alachua, Charlotte, and Santa Rosa Counties, FL. These were compared to the standard variety Emerald for a 3-yr period. Additionally, morphological differences were determined from an experiment that was managed without mowing or trimming in a three-replicate randomized complete block design conducted in Palm Beach County, FL.

Characteristics and Description

In Florida, Pristine was compared to the standard variety Emerald for eight inflorescence traits; the two varieties differed significantly for seven of these seed-head traits (Table 1). The overall inflorescence length as measured from the apex of the seed head to the uppermost or flag leaf node averaged 35.8 ± 1.17 mm for Emerald compared with $26.1 \pm$ 0.82 mm for Pristine. Pristine carried an average of 11 seeds on 9.65 \pm 0.40 mm long seed heads that were 1.22 ± 0.06

mm wide. These seed heads were significantly shorter and produced fewer seeds than Emerald (Table 1). Emerald also had a higher seed density on the raceme compared with Pristine (0.880 seed mm⁻¹ vs. 0.727 seed mm⁻¹), but this trait was not visibly discernable. In general, Pristine produced seed heads that were nearly 27% shorter and carried one-third fewer seeds than Emerald. Additionally, Pristine produced seeds that were 2.43 ± 0.08 mm in length and were significantly shorter than Emerald seeds, which measured 2.67 ± 0.06 mm. Seed of Pristine and Emerald averaged 0.81 mm and 0.89 mm in width, respectively. The overall peduncle length, as measured from the flag leaf node to the base of the seed head, and the exposed portion of the peduncle not enveloped by the sheath of the flag leaf were both significantly shorter on Pristine. Peduncle width did not differ between varieties. The aggregate measurement of these traits confirmed the visual assessment that Emerald produced a more robust

Table 1. Comparison of floral traits between zoysiagrasscultivars Emerald and Pristine.

| Inflorescence traits (in mm) | Emerald | | Pristine | | LSD |
|-----------------------------------|---------|------|----------|------|------------|
| | Mean | SE | Mean | SE | (P < 0.05) |
| Overall shoot length [†] | 35.80 | 1.17 | 26.05 | 0.82 | 5.57 |
| Seed-head length‡ | 11.70 | 0.55 | 9.65 | 0.40 | 1.97 |
| Seed-head width§ | 1.53 | 0.07 | 1.21 | 0.06 | 0.19 |
| Seed count/seed head [¶] | 16.10 | 0.87 | 10.90 | 0.48 | 3.10 |
| Exposed peduncle length# | 12.40 | 0.91 | 7.95 | 0.80 | ns |
| Peduncle length ^{††} | 23.35 | 1.36 | 15.18 | 0.51 | 5.33 |
| Glume length ^{‡‡} | 2.66 | 0.06 | 2.43 | 0.08 | 0.19 |
| Glume width§§ | 0.89 | 0.02 | 0.81 | 0.04 | 0.06 |

[†]A measure of the total inflorescence length from the apex of the seed head to the first node on the peduncle subtending the raceme.

 ${}^{\ddagger}\mbox{Measured}$ from the apex of the seed head to the top of the peduncle on the base of the raceme.

 $\$ Measured at the broadest part of the seed head.

[¶]The average number of seeds per head or raceme.

*The average length of the peduncle not covered by the flag leaf sheath.

 $^{\rm tr}{\rm The}$ average pinnacle length measured from the base of the seed head to the top of the node.

 $^{\ddagger 4}\mathrm{A}$ measure of the average glume length on the seed located at the mid point along the seed head.

^{§§}Measured at the widest part of the glume.

and coarse inflorescence, which is an aesthetically undesirable trait that detracts from turf quality.

The total number of seed heads produced in the canopy also varied by season and variety (Fig. 1). Over 12-mo establishment experiments, Pristine produced a monthly average of 3.45 ± 0.84 seed heads dm⁻², which was significantly less than the 6.40 ± 1.73 seed heads dm⁻² produced by Emerald. On an annual basis, this resulted in 46% fewer seed heads produced by Pristine than Emerald. On an individual month basis, Pristine produced seed head averages that ranged from 0 to 7.97 seed heads dm⁻², while Emerald ranged from 0 to 16.0 seed heads dm⁻² (Fig. 1). Emerald had a peak in seed-head production during February, with the



Figure 1. Comparison of the average seed head production between zoysiagrass cultivars Pristine and Emerald measured on a monthly basis in Palm Beach County, FL.

heaviest flush from December through March, and was offset from Pristine, which had a bimodal flush of seed heads produced from August through November and again in March. Emerald produced more seed heads than Pristine in 7 of 12 months, with the magnitude of the differences varying among months (Fig. 1). This result was repeated when measured on a subjective rating scale at the Charlotte County test site; Emerald again produced significantly more seed heads than Pristine in 4 of the 12 months. Similar results were obtained at the Alachua County test site, but no significant differences occurred at the northern test site in Santa Rosa County. As with a robust inflorescence, prolific production of seed heads also detracts from turf quality and appearance but further exacerbates the difficulty of managing crop purity in a clonally propagated perennial variety. In general, seed-head production is an undesirable trait in this segment of the turf industry.

Leaf texture was assessed by measurements of leaf morphology, and both varieties were again dissimilar (Table 2). Pristine exhibited shorter and narrower flag leaves as well as shorter leaf sheaths than Emerald. Stolon and leaf morphology of the vegetative tissue was measured on fully expanded leaves at the fifth node below the meristem. The leaf length of Pristine averaged 48.1 ± 2.66 mm compared with 60.8 ± 2.16 mm for Emerald. Likewise, leaf width for Pristine was 1.59 ± 0.07 mm versus 1.96 ± 0.08 mm for Emerald. In general, Pristine produced leaves that were 21% shorter and 19% narrower than Emerald. Internode length and width for Pristine averaged 20.92 ± 0.45 mm and 1.35 \pm 0.06 mm, respectively. Emerald produced internodes that averaged 11.37 ± 0.58 mm in length and 2.07 ± 0.08 mm in width. Pristine produced internodes that were 84% longer than Emerald; however, no meaningful difference existed between internode widths (Table 2). Leaf color and stolon and rhizome pigment also varied between the two varieties and was determined by a comparison of fresh leaf and sto-

lon tissue samples to color panels in the Munsell Color Chart (Munsell Color, 1977). The adaxial surface of the leaves of Pristine ranged from 7.5GY (4/6) to 7.5GY (5/6) in color. Emerald produced leaves of less-intense green color and ranged from 5GY (4/6) to 7.5GY (4/6). On rhizomes exposed to sunlight, Pristine produced a pigment that ranged up to 5RP (3/2), while Emerald produced a lighter pigment that measured 5RP (4/2). Although these color differences were not separated statistically, they were visibly discernable. A narrower and shorter leaf morphology combined with a darker green leaf color produced a canopy with better turf quality and appearance, along with a finer leaf texture. The longer internodes of Pristine did not affect turf quality but may have contributed to the rate of crop establishment.

These varieties also differed for their rate of ground coverage and crop establishment. The results from the southern test sites in Palm Beach and Charlotte County differed from Santa Rosa County on the Florida panhandle. In Palm Beach County, Pristine covered the plots faster and had

Table 2. Comparison of leaf morphology traits betweenzoysiagrass cultivars Emerald and Pristine.

| Vegetative traits (in mm) | Emerald | | Pristine | | LSD |
|------------------------------|---------|------|----------|------|------------|
| | Mean | SE | Mean | SE | (P < 0.05) |
| Flag leaf length | 9.20 | 0.99 | 4.65 | 0.44 | 1.87 |
| Flag leaf width [†] | 0.79 | 0.25 | 0.81 | 0.03 | ns |
| Flag leaf sheath length | 11.45 | 0.77 | 8.50 | 0.65 | 2.35 |
| Leaf length | 60.80 | 2.16 | 48.07 | 2.16 | 8.34 |
| Leaf width [†] | 1.96 | 0.08 | 1.59 | 0.08 | 0.25 |
| Internode length | 11.37 | 0.58 | 20.92 | 0.45 | 2.29 |
| Internode width‡ | 1.35 | 0.06 | 1.33 | 0.06 | ns |

[†]Measured at the widest part of the leaf.

[‡]Measured at the widest part of the internode.

significantly more cover in each month of the year (Fig. 2). Pristine attained full plot coverage in an average of 10.5 mo; Emerald only covered 87.5% of the prescribed area within the 12-mo period. Fifty-percent plot coverage was attained in 5.2 mo and 8.0 mo for Pristine and Emerald, respectively (Fig. 2). This result from Palm Beach County was supported and repeated in Charlotte County, where Pristine fully covered the plots in an average in 7.0 mo, and attained 50% plot coverage in about 5.3 mo. In contrast, Emerald took 10.0 mo to fully cover the plots and 6.7 mo to attain 50% coverage. At the northern test site in Santa Rosa County, there was essentially no difference in these two varieties for the rate of crop establishment. A vegetatively propagated turf variety that is established and closes canopy more rapidly is typically harvested earlier and generally requires fewer inputs. These important agronomic and economic advantages are particularly useful for growers in the southern production regions of the state, where land is more expensive, water is more restricted, and operating costs are higher.



Figure 2. Comparison of cumulative average coverage measured on a monthly basis between zoysiagrass cultivars Pristine and Emerald in Palm Beach County, FL.

In summary, Pristine differed from Emerald for four groups of traits. Horticultural traits such as leaf texture and color significantly enhanced turf quality and overall appearance. The production of fewer and smaller seed heads distributed at a lower density in the canopy imparted the appearance of a more refined floral morphology and provides both agronomic and horticultural benefits. Compared with Emerald, Pristine also exhibited a significantly faster rate of crop establishment and coverage in the southern production regions. This imparts important agronomic and economic advantages relative to harvest intervals, crop maintenance, and production costs. Taken together, Pristine exhibited a significant improvement over Emerald and should have a competitive advantage over Emerald in specific applications within the specialty zoysiagrass market.

Availability

Pristine is licensed exclusively to Environmental Turf, Inc., 4366 E. Kinsey Rd., Avon Park, FL 33825 (www.environmentalturf.com) under a master-license agreement with the Florida Foundation Seed Producers, 3913 Hwy 71, Greenwood, FL 32443. Breeder stock is maintained at the Everglades Research and Education Center, 3200 East Palm Beach Rd. IFAS-University of Florida, Belle Glade, FL 33430-8003. Breeder and Foundation stocks are also maintained by Environmental Turf under the guidelines of the Southern Seed Certification Association, Inc., P.0. Box 2619, Auburn, AL 36831 (www.ag.auburn.edu/ssca). In 2008, samples were deposited with the National Center for Genetic Resources Preservation (NCGRP), USDA-ARS, and are held at the ARS Plant Genetic Resources Conservation Unit, 1109 Experiment St., Griffin, GA 30223. Pristine was trademarked as 'PristineFlora Zoysia' (USP&TO Serial No.77154003), which is its agreed commercial designation in the United States; international nomenclature will vary depending on language or marketplace. A plant patent was submitted in 2005 under the denomination of BA-305, and approved in January, 2008 as U.S. PP 18415 P3. Unless otherwise negotiated, samples of Pristine are available for research purposes only, and request should be directed to R. T. Nagata at the Everglades Research and Education Center, IFAS/University of Florida or Environmental Turf, Inc. Sublicenses for commercial production purposes are available from Environmental Turf, Inc., and Florida Foundation Seed Producers, Inc.

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