HortScience 36(4):784–786. 2001.

Zinc Requirements of Annual Bluegrass and Creeping Bentgrass

Xia Xu[1] and Charles F. Mancino[2]
Department of Agronomy, The Pennsylvania State University, University Park, PA 16802-3504

Additional index words. *Poa annua*, *Agrostis stolonifera*, mineral nutrition, putting greens, biotypes, tissue testing, micronutrients

Abstract. **Annual bluegrass (*Poa annua* L.) is becoming an important component of golf course putting greens. A greenhouse sand culture experiment was conducted to study the zinc (Zn) requirements of three genotypes of flowering annual bluegrass (FAB) and three genotypes of vegetative annual bluegrass (VAB), which were compared with the three parents of 'Penncross' creeping bentgrass [*Agrostis stolonifera* L. (CB)]. Clonally propagated plants were grown in sand culture without Zn for 6 weeks prior to the initiation of the Zn treatments. The plants were then irrigated for 3 weeks with half-strength Hoagland's nutrient solution containing 0, 2.5, 5.0, or 40 mg·L^{-1} Zn from $ZnSO_4$. Color was the only parameter affected by genotype; each genotype showed a significant quadratic response to increasing levels of Zn, with highest color ratings occurring at 2.5 mg·L^{-1}. No genotypic differences were observed among CB, VAB, and FAB for shoot fresh and dry weight, root dry weight, or shoot tissue Zn concentrations. Shoot dry weight of all genotypes increased quadratically with Zn levels. Root dry weights of both VAB and FAB increased, while that of CB remained unchanged, as Zn level increased. Zinc concentrations in shoot tissue increased linearly as Zn level increased. Shoot Zn concentrations were higher in both VAB and FAB than in CB at each Zn level, but differences between VAB and FAB were insignificant. Maintaining shoot Zn concentrations below 109 mg·kg^{-1} in CB and 200 mg·kg^{-1} in VAB or FAB prevented Zn phytotoxicity from occurring.**

Annual bluegrass is a weed species found in many turf areas, but is so competitive it can become a major component of a turf stand. Many efforts have been made to control annual bluegrass in creeping bentgrass putting greens (Baldwin, 1993). Despite these efforts, this weed quickly invades most putting greens, and, over time, can become the predominate turf species. As a result, many golf course superintendents simply choose to manage annual bluegrass as a turf species rather than a weed on their greens (Huff, 1999).

One major problem encountered in managing annual bluegrass on greens is a lack of information regarding its nutrient requirements. Zinc (Zn) is an essential plant micronutrient that is required in the synthesis of growth hormones and proteins (Marschner, 1995). Knowing the Zn requirements of annual bluegrass will help turf managers and scientists to develop a fertilizer program to either facilitate annual bluegrass growth or to reduce its invasion into creeping bentgrass putting greens.

In general, a Zn level of 20 to 55 mg·kg^{-1} DW in shoot tissue is considered to be sufficient for turfgrasses (Jones, 1980). Boehle and Lindsay (1969) reported a Zn concentration range of 40 to 200 mg·kg^{-1}. Davis and Beckett (1977) found that a Zn concentration of 221 mg·kg^{-1} in shoot tissue of perennial ryegrass (*Lolium perenne* L.) caused phytotoxicity. However, Spear and Christians (1991) found that a Zn level of 1500 mg·kg^{-1} in shoot tissue had no deleterious effects on Penncross creeping bentgrass [*Agrostis stolonifera* L. (CB)]. 'Merlin' red fescue (*Festuca rubra* L.) was able to tolerate higher shoot tissue Zn concentrations than other red fescue cultivars, tall fescue (*Festuca arundinaceae* L.), Kentucky bluegrass (*Poa pratensis* L.), and perennial ryegrass (Chaney, 1993). Turfgrass species, cultivars, and genotypes differ in their ability to acquire or utilize Zn.

Many genotypes of annual bluegrass exist on putting greens (Mitich, 1998). Some genotypes are prolific producers of seedheads, while others flower infrequently or not at all. These annual bluegrasses belong to two subspecies: *Poa annua* sp. *annua* and *Poa annua* sp. *reptans*. The *Poa annua* sp. *annua* is a true winter annual while *Poa annua* sp. *reptans* is a short-lived perennial that makes a very good species for putting greens (Huff, 1999). However, no information is available on the Zn requirements of these annual bluegrass subspecies.

The objectives of this study were to determine the color and growth responses of different genotypes of annual bluegrasses and compare them with those of creeping bentgrass, as well as to determine if inter- and intraspecific differences exist.

Materials and Methods

To reduce the effects of genetic variation on response, three parents of the cultivar Penncross were used to represent the genotypes of creeping bentgrass. They were designated as BCB, RCB, and WCB (Table 1). The six genotypes of annual bluegrass originated from selections made from putting greens at the Oakmont Country Club in Oakmont, Pa. by David Huff (Dept. of Agronomy, The Pennsylvania State Univ., University Park, Pa.). The annual bluegrass genotypes were morphologically categorized according to flowering frequency, leaf texture, and leaf color. Genotypes 11G-2, 18G-1, and 18G-2 were categorized as frequently flowering (FAB) and genotypes 9G-1, 9G-6, and 11G-6 as infrequently flowering or vegetative annual bluegrass (VAB). Flowering frequency was measured by the percentage of major flowering tillers. Leaf texture was determined by measuring the width of the first expanded leaf. Color was visually evaluated using a rating scale of 1 to 9.

Tillers of clonally propagated plants were transplanted into containers (12.5 cm in length × 2.5 cm in diameter) filled with white silica sand (U.S. Silica, Mapleton, Pa., particle size 0.15–1.0 mm; 99.2% to 99.9% SiO_2). The plant material was established for 6 weeks using a Zn-deficient half-strength Hoagland's nutrient solution adjusted to pH 5.5–6.0

Table 1. Species and genotypes of plants used to study the effect of varying levels of Zn on color and growth of annual bluegrass and creeping bentgrass

Group	Species	Genotypes	Flowering[z]	Texture[y]	Color[x]
CB	Creeping bentgrass	BCB	NA	Medium	Green
		RCB	NA	Medium	Green
		WCB	NA	Medium	Green
VAB	Annual bluegrass	9G-1	Infrequently	Medium	Green
		9G-6	Infrequently	Medium	Dark green
		11G-6	Infrequently	Coarse	Light green
FAB	Annual bluegrass	11G-2	Frequently	Coarse	Light green
		18G-1	Frequently	Coarse	Light green
		18G-2	Frequently	Fine	Light green

[z]NA = Not applicable; infrequently flowering annual bluegrasses (VAB) have ≤10% flowering tillers; frequently flowering annual bluegrasses (FAB) have ≥50% flowering tillers.
[y]Medium texture = 1.8–2.5 mm, coarse = 2.5–3.5 mm, and fine ≤1 mm in leaf width.
[x]Color visually determined on clonal plant materials grown under identical greenhouse conditions.

Received for publication 7 Apr. 2000. Accepted for publication 7 Oct. 2000. Special thanks are due to David Huff for providing the plant material used in this study, Dianne Petrunak for technical support in this research, and Jon Chorover and Mary Kay Amistadi for the use of their analytical equipment. The cost of publishing this paper was defrayed in part by the payment of page charges. Under postal regulations, this paper therefore must be hereby marked *advertisement* solely to indicate this fact.
[1]Former Graduate Research Assistant.
[2]To whom requests for reprints should be addressed. Current address: The Scotts Company, 14111 Scottslawn Road, Marysville, OH 43041. E-mail address: charles.mancino@scottsco.com

(Hoagland and Arnon, 1950) in deionized distilled water. Greenhouse conditions were 25 °C day/15 °C night under natural light and daylengths.

After establishment, the plants were clipped to a 1- to 2-cm height and the Zn treatments were initiated. Zinc and genotype treatments were arranged in a split-plot experimental design with Zn as the main-plot factor and genotype as the subplot factor. There were four Zn treatments, nine genotypes, and four replications. Zn treatments were applied for 3 weeks by irrigating the turf every other day with 0, 2.5, 5.0, and 40 mg·kg^{-1} Zn as $ZnSO_4$. The selection of Zn treatment levels was based on the maximum allowable Zn in irrigation water (Ayers and Westcott. 1985). The plants were not clipped during the entire period of Zn treatment. The entire experiment was repeated twice.

Data were collected for leaf color, shoot and root production, and shoot tissue Zn content. Visual color ratings were taken when acute deficiency or toxicity symptoms appeared, or 3 weeks after treatment initiation. Color was visually evaluated using a rating scale of 1 to 9, with 1 = yellow, 6 = acceptable green color, and 9 = very dark green color. Shoot fresh and dry weights, and root dry weight were measured at the end of each experiment. Shoot dry weights were determined after samples were oven-dried at 80 °C for 48 h. Root dry weights were obtained after roots were carefully hand-washed in tap water and oven-dried. The dried plant tissue was then ground to pass through a 40-mesh screen (Thomas-Wiley mill, model 3383-L10; Arthur H. Thomas Scientific Apparatus, Philadelphia). Shoot tissue Zn content was determined by acid-ashing (Doty et al., 1982) and flame atomic absorption spectrophotometry (Video 22 Photometry Adsorption; Thermo Jarrell Ashe, Franklin, Mass.).

The data from the two experiments were compared to test for significant differences at each treatment level. No differences were found between the experiments, so the data from both experiments were combined and subjected to analysis of variance (ANOVA) in a split-block design using SAS ANOVA (SAS Institute, 1996). The significance of the whole plot (Zn) effect was tested using the genotype × block error term. The sub-plot (genotype) effect was determined using the genotype × Zn treatment × replication error term. Inter- or intra-specific differences within each Zn treatment level were determined using Fisher's protected LSD ($P \leq 0.05$). The response of each genotype or group of genotypes, was determined using regression analysis and the SAS general linear model (GLM) procedure (SAS Institute, 1996).

Results

Color. Each genotype showed a significant quadratic response to increasing levels of Zn (Table 2). Color was the only parameter where differences existed among genotypes within CB, VAB, and FAB. In the absence of Zn, leaf color ratings were unacceptable (<6) in two of the three parents of Penncross, and in all of the AB genotypes. Deficiency symptoms appeared as a light green color on the younger leaves of the plants. Although considered unacceptable, the VAB genotypes 9G-1 and 9G-6 had significantly higher color ratings than did VAB genotype 11G-6. The color of the latter genotype was similar to that of the FAB genotypes.

All genotypes of CB and AB had better than acceptable turf color when Zn treatment level was increased from 0 to 2.5 or 5.0 mg·L^{-1} (Table 2). The VAB genotypes 9G-1 and 9G-6 had very good color ratings (>7.0), while 11G-6 had lower color ratings that were similar to the CB genotype BCB and the FAB genotypes 11G-2 and 18G-2. Color ratings declined significantly when plants were exposed to Zn at 40 mg·L^{-1}, especially for CB genotype WCB, VAB genotype 11G-6, and FAB genotype 11G-2. The loss of turfgrass color at this high Zn concentration first began as chlorosis on younger leaves, followed by a yellowing of all leaves.

Shoot growth. No differences were observed within species for shoot fresh and dry weights. Therefore, the genotypes were grouped for further data analysis (Table 3).

Shoot fresh weight data were highly variable. Fresh weights were greater for CB and VAB than for FAB at each Zn concentration tested except 40 mg·L^{-1} (Table 3). At this high Zn level, shoot fresh weight was higher for VAB than for CB and FAB, in which values were similar. When analyzed across Zn treatment levels, only CB had a significant linear, negative response to increasing Zn levels (Table 3). The fresh weight of the AB biotype was not significantly affected by Zn treatment level because of the large variability in shoot fresh weights.

The variability observed in shoot fresh weight was not as evident for shoot dry weight (Table 3). A quadratic growth response with increasing Zn levels was measured for all the plant materials tested. Highest shoot dry weights occurred at 2.5 and 5.0 mg·L^{-1} Zn. Within every Zn treatment level except 40 mg·L^{-1}, CB produced the highest shoot dry weight, followed by VAB and then FAB. Within the 40 mg·L^{-1} Zn treatment level, CB shoot dry weight declined considerably and became similar to that of VAB. At this high concentration, shoot dry weight of CB was lower than that of the untreated controls. Although dry weight of VAB and FAB also declined at this high level of Zn, it became about equal to that of the untreated controls.

Root production. Creeping bentgrass produced more root dry matter than did VAB and FAB at all Zn levels tested (Table 3); the latter biotypes were similar to one another in root production. However, there appeared to be a trend for greater root production with VAB than for FAB. Root production increased linearly as Zn treatment level increased for the AB biotypes, but CB root production remained the same.

Zn concentration in shoots. Concentrations of Zn in shoot tissue differed between the two species. Both VAB and FAB had significantly higher Zn concentrations than did CB at every Zn level tested (Table 3). No differences between VAB and FAB were detected. Tissue Zn concentration increased linearly in all plant material as Zn treatment level increased (Table 3).

Discussion

Very little research has been performed on the Zn requirements of CB and AB. Moreover, no literature exists that compares the Zn requirements of vegetative and flowering biotypes of AB. Jones (1980) reported a general Zn sufficiency range of 20 to 55 mg·kg^{-1} in turfgrass shoot tissue, but Waddington and Zimmerman (1972) reported higher average concentrations of 78 and 61 mg·kg^{-1} in field-grown AB and CB, respectively. In our study, shoot tissue Zn concentrations below ≈20 mg·kg^{-1} in CB and AB were not adequate for acceptable turf color and shoot growth; this is in agreement with the data of Jones (1980). However, our sufficiency levels of Zn in shoot tissue for acceptable green color were ≈145 mg·kg^{-1} in AB and 74 mg·kg^{-1} in CB, which are higher than the levels reported by Waddington and Zimmerman (1972), perhaps because they reported average Zn concentrations for an entire growing season. They had reported that leaf tissue Fe concentrations in colonial bentgrass (*Agrostis capillaris* L.) could average 204 mg·kg^{-1}, but ranged from 106 to 362 mg·kg^{-1} depending upon time of year.

Table 2. Genotypic leaf color[z] differences for three creeping bentgrasses (CB), three vegetative annual bluegrasses (VAB), and three flowering annual bluegrasses (FAB) in response to varying levels of Zn.

		Zn treatment (mg·L^{-1})				
Group	Genotype	0	2.5	5.0	40	Regression
CB	BCB	5.6 a–c[y]	6.1 d	6.2 b	5.2 a	Q***
	RCB	6.1 a	6.7 b	6.7 a	5.3 a	Q***
	WCB	5.4 b–d	6.5 bc	6.4 ab	3.6 e	Q***
VAB	9G-1	5.7 a–c	7.1 a	6.8 a	5.1 ab	Q***
	9G-6	5.9 ab	7.3 a	6.8 a	4.6 bc	Q***
	11G-6	5.0 d	6.1 d	6.0 b	4.0 de	Q***
FAB	11G-2	5.3 cd	6.4 b–d	6.1 b	3.8 e	Q***
	18G-1	5.2 cd	6.5 bc	6.1 b	4.4 cd	Q***
	18G-2	5.3 cd	6.3 cd	6.1 b	4.5 cd	Q***

[z]Color was visually rated on a scale of 1 to 9 with 1 = brown turf, 6 = acceptable green color, and 9 = very dark green color. Data are the averages for eight replications.
[y]Mean separation within columns by LSD ($P \leq 0.05$).
[***]Significant quadratic response at $P \leq 0.001$.

Table 3. Shoot and root weights, and shoot tissue Zn concentration[z] of creeping bentgrass (CB), vegetative annual bluegrass (VAB), and flowering annual bluegrass (FAB) in response to varying levels of Zn.

	Zn treatment (mg·L^{-1})				
Group	0	2.5	5.0	40	Regression
			Shoot fresh weight (g)		
CB	6.0 a	5.7 ab	5.8 a	4.3 b	L***
VAB	6.1 a	6.1 a	6.4 a	5.3 a	NS
FAB	4.1 b	5.0 b	5.0 b	4.3 b	NS
			Shoot dry weight (g)		
CB	1.6 a	1.6 a	1.6 a	1.3 a	Q***
VAB	1.2 b	1.2 b	1.2 b	1.2 a	Q***
FAB	0.9 c	1.1 c	1.0 c	0.9 b	Q***
			Root dry weight (g)		
CB	2.0 a	2.4 a	2.2 a	2.0 a	NS
VAB	1.1 b	1.2 b	1.4 b	1.5 b	L***
FAB	1.0 b	1.1 b	1.2 b	1.4 b	L***
			Tissue Zn (mg·kg^{-1})		
CB	14.2 a	73.5 a	108.6 a	501.4 a	L***
VAB	18.6 b	147.2 b	203.3 b	1080.2 b	L***
FAB	18.7 b	142.3 b	198.0 b	1074.7 b	L***

[z]Each number is the mean for three genotypes.
[y]Mean separation within parameters and columns by $LSD_{0.05}$.
NS, *, **, ***Nonsignificant or significant at $P \leq 0.05$, 0.01, and 0.001, respectively. L = linear response; Q = quadratic response.

Therefore, the concentrations we report in our study for Zn are probably within the range that these authors found. We also found that both VAB and FAB have similar Zn concentrations in shoot tissue, and that these concentrations are higher than those found in CB exposed to similar levels of Zn.

Color and shoot production changed very little as Zn treatment levels increased from 2.5 to 5.0 mg·L^{-1}. Turner (1993) reported no response of 'Penncross' CB to increasing levels of Zn, and Deal and Engel (1965) reported no change in Kentucky bluegrass sod color and top growth when treated with Zn. However, Deal and Engel (1965) did report an increase in root production of Kentucky bluegrass sod as Zn treatment levels increased. Wu et al. (1981) described no decline in the root production of two hybrid bermudagrasses (*Cynodon dactylon* L. ×*transvaalensis* Burtt-Davy) exposed to Zn at 50 mg·L^{-1}. Mills and Jones (1996) found that leaf tissue Zn concentrations above 200 mg·kg^{-1} could result in Zn toxicity symptoms. We found no change in the root production of CB at Zn concentrations up to 40 mg·L^{-1}, while response of VAB and FAB to increasing Zn levels was positive and linear. Therefore, CB and AB appear to have different root growth responses to Zn, while VAB and FAB have similar responses. Also, the negative effects of high levels of Zn on both CB and AB appear to be confined to the shoots. Chlorosis from high Zn levels could be an induced deficiency of magnesium or iron because their ion radii are similar to that of Zn (Chaney, 1993).

Based on our color and growth data, shoot tissue Zn concentrations that were associated with the best combination of CB, VAB, and FAB were 73.0 to 109.0, 147.2 to 203.3, and 142.3 to 198.0 mg·kg^{-1}, respectively. Shoot tissue Zn concentrations above these ranges could result in reduced turf color and growth.

Literature Cited

Ayers, R.S. and D.W. Westcott. 1985. Water quality for agriculture. Food and Agr. Org. Irrig. and Drainage Paper #29, Rome.

Baldwin, N.A. 1993. Chemical control of *Poa annua*: A review. J. Sports Turf Res. Inst. 69:7–17.

Boehle, J., Jr. and W.L. Lindsay. 1969. Micronutrients—The fertilizer shoe-nail. Part 6, In the limelight—Zinc. Fert. Solutions 13:6–12.

Chaney, R.L. 1993. Zn phytotoxicity, p. 135–161. In: A.D. Robson (ed.). Zinc in soils and plants. Kluwer Academic, Dordrecht, The Netherlands.

Davis, R.D. and P.H.T. Bechett. 1977. Upper critical levels of toxic elements in plants. New Phytol. 80:2332.

Deal, E.D. and R.E. Engel. 1965. Iron, manganese, boron, and zinc: Effects on growth of Merion Kentucky bluegrass. Agron. J. 57:533–555.

Doty, W.T., M.C. Amacher, and D.E. Baker, 1982. Manual of methods. Soil and environmental chemistry laboratory. The Pennsylvania State Univ. Inform. Rpt. 121:1–14.

Hoagland, D.H. and D.I. Arnon. 1950. The water culture method for growing plants without soil. California Agr. Expt. Sta. Circ. 347:4–31.

Huff, D.R. 1999. For richer, for *Poa*: Cultivar development of green-type *Poa annua*. U.S. Golf Assn. Green Section Record (Jan./Feb.) 11–14.

Jones, J.B. 1980. Turf analysis. Golf Course Mgt. 48(1):29–32.

Marschner, H. 1995. Functions of mineral nutrients: Micronutrients, p. 347–364. In: H. Marschner (ed.). Mineral nutrition in higher plants. 2nd ed. Academic, New York.

Mills, H.A. and J.B. Jones Jr. 1996. Plant analysis handbook II. MicroMacro Publ., Athens, Ga.

Mitich, W.L. 1998. Annual bluegrass (*Poa annua* L.). Weed Technol. 12: 414–416.

SAS Institute. 1996. SAS/STAT guide for personal computers, vers. 6.12. SAS Institute, Cary, N.C.

Spear G.T. and N.E. Christians. 1991. Creeping bentgrass response to zinc in modified soil. Commun. Soil Sci. Plant Anal. 22:2005–2016.

Turner, T.R. 1993. Turfgrass, p. 187–196. In: W.F. Bennett (ed.). Nutrient deficiencies and toxicities in crop plants. Amer. Phytopathol. Soc. Press, St. Paul, Minn.

Waddington, W.V. and T.L. Zimmerman. 1972. Growth and chemical composition of eight turfgrasses grown under high water table conditions. Commun. Soil Sci. Plant Anal. 3:329–337.

Walley, K.A., M.S.I. Khan, and A.D. Bradshaw. 1974. The potential for evolution of heavy metal tolerance in plants. I. Copper and zinc tolerance in *Agrostis tenuis*. Heredity 32:309–319.

Wu, L., D.R. Huff, and J.M. Johnson. 1981. Metal tolerance of bermudagrass cultivars, p. 35–40. In: R.W. Sheard (ed.). Proc. 4th Intl. Turfgrass Res. Conf. 4:35–40, Univ. of Guelph Press, Guelph, Ont., Canada.