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Silicon Enhances Antioxidant Capacity and Photochemical Efficiency in Drought-Stressed Creeping Bentgrass (*Agrostis stolonifera* L.) Putting Greens

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Abstract: Creeping bentgrass (*Agrostis stolonifera* L.) is an important cool-season turfgrass species that is not well understood. The objective of this study was to determine the effects of the mechanisms underlying silicon (Si) on creeping bentgrass drought tolerance under field conditions from 2022 to 2023. Five treatments, including a control (potassium silicate at 0.95 and 1.90 mL m^{-2}), Dyamin-OSA at 0.64 and 1.28 mL m^{-2}, and Agsil 21 at 0.35 mL m^{-2}, were arranged in a randomized block design with four replications and applied biweekly to creeping bentgrass putting greens during summer months. Deficit irrigation was applied to induce drought stress in June and July. The Si treatments exhibited beneficial effects on turf quality, physiological fitness, and root viability. K-silicate at 1.90 mL m^{-2} and Agsil 21 at 0.35 mL m^{-2} increased the leaf Si content by 32.0% and 22.8%, respectively, when compared to the control, as measured at the end of the trial. Among the treatments, K-silicate at 1.90 mL m^{-2}, Dyamin-OSA at 0.64 mL m^{-2}, and Agsil 21 at 0.35 mL m^{-2} tended to have greater beneficial effects than other Si treatments. Exogenous Si may improve drought tolerance by enhancing root growth and viability, Si uptake by roots, and antioxidant capacity and by protecting photosynthetic function.

Keywords: biostimulant; chlorophyll; drought; root viability; Silicon; superoxide dismutase; turfgrass

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1. Introduction

Creeping bentgrass is a dominant cool-season turfgrass species used for golf course putting greens and tees; however, it suffers summer stress-induced quality decline in the U.S. transition zone and other regions with similar climates [1,2]. Turfgrass quality decline is due to drought and heat stress during the summer [1–3]. Various agronomic and chemical approaches have been used to alleviate quality decline in creeping bentgrass during summer months. Plant biostimulants have been widely used for sustainable turf management. Certain inorganic elements, such as silicon (Si) and chelated iron, have been used as biostimulants to improve turfgrass abiotic stress tolerance and performance.

Silicon is not considered to be an essential nutrient for plants; however, it is often called a "quasi-essential" element that is not strictly needed for basic life functions but provides significant benefits to plant growth and development, especially in stress conditions. Some plants cannot complete their lifecycle in the absence of Si [2,4]. Si improves plant tolerance to various abiotic and biotic stresses [5]. Different forms of Si have varying effects on plants. Some forms of Si, such as silica, silica nanoparticles, silica gel, and potassium silicate, have been shown to improve plant growth and protect plants against diseases, while other forms, like calcium silicate and monosilicic acid [Si(OH)$_4$], may improve plant growth and tolerance to abiotic stress [2,6]. Si exists in soil solutions in the form of monosilicic acid (Si[OH]$_4$), and the Si concentration in soil solutions was reported to be from 0.4 to 2000 μmol L^{-1}, with the average ranging from 100 to 600 μmol L^{-1} [5,7]. Si is absorbed via roots in the form of H_4SiO_4, either passively via mass flow or actively via an NIP2 transporter [8]. The Si concentration in plants ranges from 0.005 to 10 mg kg^{-1} [9]. Under abiotic stress conditions, such as drought and heat, the metabolic energy of plants is limited because of the reduction in photosynthetic activity, and this may cause a reduction in Si uptake from soil solutions, because the energy required for Si uptake and assimilation is limited. Foliar application of Si may help plants to uptake and assimilate it to maintain optimum Si concentration in plants, especially under abiotic stress [2]. Liner increases in leaf tissue Si was found as the Si application rate increased [10]. However, no change in leaf Si concentration was observed in response to exogenous Si in creeping bentgrass in another study [10]. It appears that exogenous Si may influence plants by a not-yet well-defined mechanism.

It was reported that exogenous Si improved turfgrass tolerance to abiotic stress [5,11]; however, the mode of action of Si's effect on plant tolerance to abiotic stress is not well understood, especially under field conditions. Several researchers have summarized the role of Si in mitigating drought stress-induced injury to plants [5,12,13]. Eneji et al. (2008) [6] reported that three sources of Si (silica gel, $CaSiO_3$, and K_2SiO_3) applied at 1 g kg^{-1} to sandy soil increased dry matter, with potassium silicate being more effective than calcium silicate in tall fescue under deficit irrigation conditions. Bae et al. (2017) [14] found that Si application increased the root biomass of Kentucky bluegrass under drought stress conditions. A recent study by Zhang et al. (2024) [2] showed that the application of Si to creeping bentgrass may improve drought and heat stress tolerance associated with increased root growth and antioxidant metabolism under controlled environments. However, these studies were conducted in a controlled environment, and data under field conditions are lacking, especially for the turfgrasses for golf course putting greens under field conditions in the U.S. transition zone.

Abiotic stress may damage plants through an over accumulation of reaction oxygen species (ROS) and thus oxidative injury to plant cells [2]. Plants have developed various antioxidant systems to scavenge ROS and protect cells under abiotic stress [15]. Previous studies showed that exogenous Si may enhance antioxidant metabolism [11,16]. Schmidt et al. (1999) [16] reported that potassium silicate treatments improved photochemical efficiency, chlorophyll content, root and shoot growth, and antioxidant SOD activity in creeping bentgrass under drought stress. A similar result was found with creeping bentgrass by Merewitz and Liu (2019) [11]. The results from a meta-analysis of 145 experiments indicate that Si can effectively alleviate oxidative stress but was not always consistent in the effects of antioxidants within the plant leaves [17]. Zhang et al. (2024) indicated that SOD activity is responsive to Si treatments in creeping bentgrass. Si may [2] accumulate in cell walls to stabilize the cells under stress, and reduced cell electrolyte leakage improved drought and heat stress tolerance [11,18].

Although Si is not considered to be an essential macronutrient, the content of Si in some plants can be in the range of certain macronutrients. These Si deposits are commonly found on leaf and stem hairs and on the outer epidermal walls, which benefits plant tolerance to drought. However, previous studies were conducted in controlled environments, and the leaf Si content was not analyzed. We hypothesized that exogenous Si may increase root growth to facilitate Si uptake and accumulation in leaf tissues and also enhance leaf antioxidant enzyme SOD activity to protect photosynthetic function and pigment in creeping bentgrass putting greens during summer months. The objective of this study was to determine the effects and the mechanisms underlying Si products on turf quality, physiological fitness, root growth characteristics, and root viability in creeping bentgrass under field conditions.

2. Materials and Methods

2.1. Site Description and Si Treatments

This study was conducted on the 'A4' creeping bentgrass putting greens at Virginia Tech Turfgrass Research Center in Blacksburg, VA, from 8 June to 31 August 2022. The trial was repeated from 8 June to 31 August 2023. The trial included 6 treatments with 4 replications. The plot size was 1.524 × 1.829 m. Regular mowing at 9.5 mm and daily irrigation were performed. The Si products, including potassium silicate with 4.7% silica (SiO_2) (Harrell's LLC, Lakeland, FL, USA), Dyamin-OSA with 2% orthosilicic acid [$Si(OH)_4$] (plant available Si at 0.6%) from Dyacare (Bournemouth, BH1 1JU, UK), and Agsil 21 with 26.5% silica (SiO_2) from Certis Biologicals (Columbia, MD, USA), were used in this study.

Treatments included the following: 1. A fertilized control (0.73 g N m^{-2}), 2. K-silicate (0.95 mL m^{-2}), 3. K-silicate (1.90 mL m^{-2}), 4. Dyasil-OSA Si (0.64 mL m^{-2}), 5. Dyamin-OSA Si (1.28 mL m^{-2}), and 6. Agsil 21 (0.35 mL m^{-2}). A randomized block design was used with four replications. The products were dissolved in water and applied at 81.5 mL m^{-2} biweekly. The grass was fertilized with a complete fertilizer (28N-8P-18K) with micronutrients biweekly. The rate of N fertilization for treatments #2, 3, 4, 5, and 6 was the same as treatment #1 (fertilized control), so all six treatments received identical N input. In addition, the potassium rate from K-silicate and other treatments remained the same.

2.2. Drought Stress Treatment

The trials lasted for 12 weeks from 8 June to 31 August, and there was a total of six treatment applications. From 21 June to 30 June and from 11 July to 20 July, irrigation was withheld to allow for a gradual decline in the soil water content, inducing a decline in the leaf color and turf quality.

2.3. Measurements

At day 0, 14, 28, 42, 56, 70, and 84 following the first application of Si, the following parameters were measured. Leaf samples were collected at each measurement date, frozen with liquid nitrogen, and stored at −80 °C for the analysis of various metabolites (chlorophyll and carotenoids) and antioxidant enzyme SOD activity. At the same time, a small subsample from each plot was collected and dried at 65 °C and ground into a powder for analysis of leaf Si content. At the beginning (22 June and 12 July), mid (26 June and 16 July), and end (30 June and 20 July) of each dry-down cycle, additional measurements, including turf quality, leaf color, and PE, were performed. The irrigation was resumed at the end of each dry-down cycle.

2.4. Turf Quality

Turf quality was rated on a visual scale from 1 to 9, with 1 indicating completely dead or brown leaves, 6 representing minimum acceptability, and 9 indicting turgid and green leaves, with optimum canopy uniformity and density [1,19].

2.5. Photochemical Efficiency (PE)

PE was measured with a chlorophyll fluorometer (Mini Pam II, photosynthetic analyzer, Walz Photosynthesisi Instruments, Effeltrich, Germany) based on the Fv/Fm, according to the method by Zhang et al. (2024) [2].

2.6. Leaf Pigment Content

Frozen leaf tissues were ground into a powder, and chlorophyll and carotenoids were extracted with acetone. The chlorophyll and carotenoids were measured with a spectrophotometer and calculated based on a formula described by Zhang et al. [1,2]. The formulas used for chlorophyll and total carotenoid calculation are as follows:

Chl a (μg mL) = (11.24 × A661.6 nm) − (2.04 × 644.8 nm).

Chl b (μg mL) = (20.13 × A644.8 nm) − (4.19 × A661.6 nm).

Chl a + b (μg/mL) = (7.05 × A661.1 nm) + (18.09 × A644.8 nm).

Carotenoids (μg mL) = [(1000 × A470 nm) − (1.90 × chl a − 63.14 × chl b)].

2.7. Leaf Antioxidant Superoxide Dismutase (SOD) Activity and H_2O_2 Content

Leaf samples were collected biweekly, and the SOD activity was analyzed according to the method as described by Wu et al. (2017) [15]. The leaf H_2O_2 content was measured according to the procedure by Bernt and Bergmeyer (1974) [20], as described by Zhang et al. (2024) [2].

2.8. Leaf Si Concentration

The leaf Si content was extracted and analyzed according to the method by Elliott and Snyder (1991) [21] and Kraska and Breitenbeck (2010) [22], with minor modifications. Briefly, the dried leaf sample was ground and made wet with 50% H_2O_2 in a 50 mL tube, to which 50% NaOH was added before autoclaving at 138 kPa for 1 h. The silicon concentration was determined according to the method by Elliott and Snyder (1991) [21] and Kraska and Breitenbeck (2010) [22].

2.9. Root Growth Characteristics and Viability

On 31 August, four root samples (four cores with a 1.9 cm diameter and 15.24 cm deep) were collected from each plot. Root growth characteristics (root length, root diameter, root surface area, and root volume) were analyzed using the WinRhizo software (Version Pro) after the roots were completely cleaned [2]. The root dry weight was determined after the samples were dried at 65 °C for 72 h. Root viability was determined following the method described by Zhang et al. (2022) [1].

2.10. Experimental Design and Statistical Analysis

In both 2022 and 2023, a randomized block design was used with four replicates. Since no interactions for all measurements were found between the two years, the results from 2022 and 2023 were pooled, and the data were analyzed with a one-way analysis of variance (ANOVA) following the general linear model using SAS (version 9.4 for Window; SAS Institute, Cary, NC, USA, 2016). The six treatments were compared by using Fisher's protected least significant difference test at $p = 0.05$.

3. Results

3.1. Turf Quality

Foliar application of Si products improved the turf quality, beginning on 26 June (Table 1). K-silicate at 1.90 mL m^{-2}, Dyamin-OSA at 0.64 mL m^{-2}, and AgSil 21 at 0.35 mL m^{-2} had higher turf quality ratings in 6 out 9 ratings, 5 out of 9 ratings, and 7 out of 9 ratings when compared to the control, as measured from 26 June to 31 August. At the end of the 2nd dry-down cycle (20 July), K-silicate at 1.9 mL m^{-2}, Dyamin-OSA at 0.64 and 1.28 mL m^{-2}, and Agsil 21 at 0.35 mL m^{-2} increased turf quality ratings by 5.8%, 8.7%, 7.2%, and 10.1%, respectively, when compared to the control.

Table 1. Turf quality responses to foliar application of three silicon sources in creeping bentgrass putting greens.

Treatment	Rate	Turf Quality (1–9, 9 = Best)					
	(mL m^{-2})	8 June	22 June	26 June	30 June	6 July	12 July
1. Control	0	7.5 a	6.7 a	6.3 c	6.3 b	6.7 b	7.4 b
2. K-silicate	0.95	7.5 a	6.9 a	6.5 bc	6.5 ab	7.0 ab	7.6 ab
3. K-silicate	1.9	7.3 a	6.8 a	6.7 abc	6.9 a	7.2 a	7.8 a
4. Dyamin-OSA	0.64	7.5 a	7.0 a	7.1 a	6.9 a	7.2 a	7.7 a
5. Dyamin-OSA	1.28	7.6 a	6.7 a	6.8 ab	6.7 ab	7.0 ab	7.9 a
6. Agsil 21	0.35	7.5 a	6.8 a	6.7 abc	6.9 a	7.2 a	7.9 a
		16 July	20 July	3 August	17 August	31 August	
1. Control	0	6.8 b	6.9 c	6.5 a	6.5 b	6.8 b	
2. K-silicate	0.95	7.1 ab	7.1 bc	6.6 a	7.0 a	7.2 ab	
3. K-silicate	1.9	7.1 ab	7.3 ab	6.7 a	7.0 a	7.2 ab	
4. Dyamin-OSA	0.64	7.2 ab	7.5 a	6.6 a	6.9 ab	7.1 ab	
5. Dyamin-OSA	1.28	7.2 ab	7.4 ab	6.9 a	7.1 a	7.3 a	
6. Agsil 21	0.35	7.4 a	7.6 a	7.0 a	7.2 a	7.3 a	

Means followed by the same letters within the same column are not significantly different at $p = 0.05$, using Fisher's protected least significant difference test.

3.2. Leaf Photochemical Efficiency (PE)

Foliar application of Si products improved the leaf PE, beginning on 30 June (Table 2). K-silicate at 1.90 mL m^{-2}, Dyamin-OSA at 0.64 mL m^{-2}, and AgSil 21 at 0.35 mL m^{-2} had greater PE values in 6 out 8 ratings, 5 out of 8 ratings, and 3 out of 9 ratings when compared to the control, as measured from 30 June to 31 August. At the end of the 2nd dry-down cycle (20 July), K-silicate at the two rates, Dyamin-OSA at 1.28 mL m^{-2}, and Agsil 21 increased the PE by 15.2%, 15.2%, 14.5%, and 8.8%, respectively, when compared to the control.

Table 2. Leaf photochemical efficiency (PE) response to foliar application of three silicon sources in creeping bentgrass putting greens.

Treatment	Rate	PE (Fv/Fm)					
	(mL m^{-2})	8 June	22 June	26 June	30 June	6 July	12 July
1. Control	0	0.761 a	0.711 a	0.625 a	0.612 b	0.736 a	0.720 b
2. K-silicate	0.95	0.778 a	0.736 a	0.731 a	0.725 a	0.742 a	0.769 a
3. K-silicate	1.90	0.739 a	0.737 a	0.716 ab	0.727 a	0.737 a	0.743 a
4. Dyamin-OSA	0.64	0.741 a	0.729 a	0.715 ab	0.707 a	0.737 a	0.772 a
5. Dyamin-OSA	1.28	0.812 a	0.736 a	0.742 a	0.743 a	0.745 a	0.765 a
6. Agsil 21	0.35	0.735 a	0.776 a	0.738 a	0.726 a	0.747 a	0.779 a
		16 July	20 July	3 August	17 August	31 August	
1. Control	0	0.621 bc	0.581 b	0.699 c	0.757 b	0.735 c	
2. K-silicate	0.95	0.709 ab	0.669 a	0.816 ab	0.865 a	0.791 ab	
3. K-silicate	1.90	0.716 ab	0.669 a	0.799 ab	0.869 a	0.815 a	
4. Dyamin-OSA	0.64	0.678 bc	0.614 ab	0.831 a	0.867 a	0.798 a	
5. Dyamin-OSA	1.28	0.711 ab	0.665 a	0.809 ab	0.867 a	0.790 ab	
6. Agsil 21	0.35	0.740 a	0.632 ab	0.741 bc	0.825 ab	0.785 abc	

Means followed by the same letters within the same column are not significantly different at $p = 0.05$, using Fisher's protected least significant difference test.

3.3. Leaf Chlorophyll Content

Foliar applications of all Si products increased the leaf Chl content when compared to the control from 20 July to 31 August, except for K-silicate at the lower rate on 3 August and 31 August and Dyamin-OSA at the higher rate (Table 3). At the end of the 2nd dry-down cycle (20 July), K-silicate at 0.95 and 1.9 mL m^{-2}, Dyamin-OSA at 0.64 and 1.28 mL m^{-2}, and Agsil 21 at 0.35 mL m^{-2} increased the leaf chlorophyll content by 24.4%, 25.8%, 21.1%, 20.2%, and 23.5%, respectively, when compared to the control.

Table 3. Leaf chlorophyll (Chl) content response to foliar application of three silicon sources in creeping bentgrass putting greens.

Treatment	Rate	Chl (mg/g FW)			
	(mL m^{-2})	8 June	22 June	6 July	20 July
1. Control	0	2.48 a	3.08 a	2.75 a	2.13 b
2. K-silicate	0.95	2.35 a	3.26 a	3.46 a	2.65 a
3. K-silicate	1.90	2.24 a	3.13 a	3.27 a	2.68 a
4. Dyamin-OSA	0.64	2.49 a	3.04 a	3.46 a	2.58 a
5. Dyamin-OSA	1.28	2.23 a	3.03 a	3.39 a	2.56 a
6. Agsil 21	0.35	2.43 a	2.86 a	3.36 a	2.63 a
		3 August	17 August	31 August	
1. Control	0	2.89 b	2.31 b	2.22 b	
2. K-silicate	0.95	3.20 ab	2.66 a	2.58 ab	
3. K-silicate	1.90	3.40 a	2.73 a	2.66 a	
4. Dyamin-OSA	0.64	3.35 a	2.85 a	2.63 a	
5. Dyamin-OSA	1.28	3.33 a	2.88 a	2.57 ab	
6. Agsil 21	0.35	3.51 a	2.76 a	2.66 a	

Means followed by the same letters within the same column are not significantly different at $p = 0.05$, using Fisher's protected least significant difference test.

3.4. Leaf Carotenoids Content

Foliar application of all Si products increased the leaf carotenoid content when compared to the control from 20 July to 17 August (Table 4). At the end of the trial, K-silicate at the higher rate, Dyamin-OSA at the lower rate, and Agsil 21 treatments increased the carotenoid content when compared to the control. At the end of the 2nd dry-down cycle (20 July), K-silicate at 0.95 and 1.9 mL m^{-2}, Dyamin-OSA at 0.64 and 1.28 mL m^{-2}, and Agsil 21 at 0.35 mL m^{-2} increased the leaf carotenoid content by 21.0%, 27.2%, 23.5%, 22.2%, and 23.5%, respectively, when compared to the control.

Table 4. Leaf carotenoid content response to foliar application of three silicon sources in creeping bentgrass putting greens.

Treatment	Rate	Carotenoids (mg/g FW)			
	(mL m^{-2})	8 June	22 June	6 July	20 July
1. Control	0	0.85 a	1.32 a	1.08 a	0.81 b
2. K-silicate	0.95	0.80 a	1.35 a	1.35 a	0.98 a
3. K-silicate	1.90	0.76 a	1.32 a	1.29 a	1.03 a
4. Dyasil-OSA	0.64	0.85 a	1.24 a	1.36 a	1.00 a
5. Dyasil-OSA	1.28	0.76 a	1.22 a	1.32 a	0.99 a
6. Agsil 21	0.35	0.83 a	1.17 a	1.31 a	1.00 a
		3 August	17 August	31 August	
1. Control	0	1.05 b	0.92 b	0.85 b	
2. K-silicate	0.95	1.21 ab	1.08 a	1.00 ab	
3. K-silicate	1.90	1.28 ab	1.11 a	1.04 a	
4. Dyamin-OSA	0.64	1.27 ab	1.13 a	1.04 a	
5. Dyamin-OSA	1.28	1.35 a	1.14 a	1.00 ab	
6. Agsil 21	0.35	1.34 a	1.10 a	1.03 a	

Means followed by the same letters within the same column are not significantly different at $p = 0.05$, using Fisher's protected least significant difference test.

3.5. Leaf SOD Activity

Foliar application of K-silicate at 1.90 mL m^{-2} improved leaf SOD activity when compared to the control, as measured from 22 June to 31 August (Table 5). Dyamin-OSA at 1.28 mL m^{-2} and Agsil at 0.35 mL m^{-2} also improved SOD activity when compared to the control, as measured on 6 July, 20 July, 17 August, and 31 August. At the end of the 2nd dry-down cycle (20 July), K-silicate at 0.95 and 1.9 mL m^{-2}, Dyamin-OSA at 0.64 and 1.28 mL m^{-2}, and Agsil 21 at 0.35 mL m^{-2} increased leaf SOD activity by 20.6%, 21.5%, 21.0%, 23.8%, and 25.7%, respectively, when compared to the control.

Table 5. Leaf superoxide dismutase (SOD) activity response to foliar application of three silicon sources in creeping bentgrass putting greens.

Treatment	Rate	Leaf SOD Activity (Unit/g FW)			
	(mL m^{-2})	8 June	22 June	6 July	20 July
1. Control	0	789.7 a	793.1 b	695.3 b	726.9 b
2. K-silicate	0.95	803.4 a	841.4 ab	758.8 ab	876.8 a
3. K-silicate	1.90	769.8 a	936.1 a	781.2 a	883.3 a
4. Dyamin-OSA	0.64	801.4 a	796.2 b	760.5 ab	879.6 a
5. Dyamin-OSA	1.28	799.7 a	887.6 ab	806.7 a	900.1 a
6. Agsil 21	0.35	797.3 a	893.5 ab	790.1 a	913.4 a
		3 August	17 August	31 August	
1. Control	0	649.1 c	696.5 b	723.1 b	
2. K-silicate	0.95	865.4 ab	825.6 a	804.3 ab	
3. K-silicate	1.90	890.1 a	901.4 a	832.3 a	
4. Dyamin-OSA	0.64	703.4 c	893.1 a	796.3 ab	
5. Dyamin-OSA	1.28	750.3 bc	873.2 a	829.6 a	
6. Agsil 21	0.35	717.7 c	851.0 a	848.7 a	

Means followed by the same letters within the same column are not significantly different at $p = 0.05$, using Fisher's protected least significant difference test.

3.6. Leaf H_2O_2 Content

Foliar application of the three Si sources reduced the leaf H_2O_2 content when compared to the control, as measured from 6 July to 31 August, except for K-silicate at 0.95 mL m^{-2} and Dyamin-OSA at 0.64 mL m^{-2} on 17 August (Table 6). At the end of the 2nd dry-down cycle (20 July), K-silicate at 0.95 and 1.90 mL m^{-2}, Dyamin-OSA at 0.64 and 1.28 mL m^{-2} and Agsil 21 at 0.35 mL m^{-2} reduced the leaf H_2O_2 content by 10.3%, 17.6%, 21.6%, 18.1%, and 23.7%, respectively, when compared to the control.

Table 6. Leaf hydrogen peroxide (H_2O_2) content response to foliar application of three silicon sources in bentgrass putting greens.

Treatment	Rate	H_2O_2 Content (μmol g^{-1} FW)			
	(mL m^{-2})	8 June	22 June	6 July	20 July
1. Control	0	22.0 a	24.2 a	36.1 a	52.4 a
2. K-silicate	0.95	22.2 a	22.7 b	31.8 c	47.0 b
3. K-silicate	1.9	22.5 a	23.3 ab	30.5 cd	43.2 bc
4. Dyamin-OSA	0.64	22.7 a	23.0 ab	31.2 cd	41.1 c
5. Dyamin-OSA	1.28	21.5 a	23.3 ab	33.8 b	42.9 bc
6. Agsil 21	0.35	21.7 a	22.8 b	29.7 d	40.0 c
		3 August	17 August	31 August	
1. Control	0	46.3 a	40.6 a	40.8 a	
2. K-silicate	0.95	40.5 bc	39.0 ab	37.4 b	
3. K-silicate	1.9	38.5 c	36.7 c	34.9 c	
4. Dyamin-OSA	0.64	42.7 b	39.1 ab	37.4 b	
5. Dyamin-OSA	1.28	41.8 b	37.5 bc	34.2 c	
6. Agsil 21	0.35	41.8 b	38.0 bc	34.1 c	

Means followed by the same letters within the same column are not significantly different at $p = 0.05$.

3.7. Leaf Si Content

Foliar application of the Si products improved the leaf Si concentration when compared to the control, as measured on 3 August, 17 August, and 31 August (Table 7). K-silicate at 1.90 mL m^{-2} and Agsil 21 at 0.35 mL m^{-2} consistently increased the leaf Si content by when compared to the control, as measured from 3 August to 31 August. At the end of the trial, K-silicate at 1.90 mL m^{-2} and Agsil 21 at 0.35 mL m^{-2} increased the leaf Si content by 32.0% and 23.83%, respectively, when compared to the control.

Table 7. Leaf silicon content response to foliar application of three silicon sources in creeping bentgrass putting greens.

Treatment	Rate	Si Content (mg/g DW)			
	(mL m^{-2})	8 June	22 June	6 July	20 July
1. Control	0	1.24 a	1.24 a	1.18 ab	1.24 a
2. K-silicate	0.95	1.16 a	1.23 a	1.22 ab	1.30 a
3. K-silicate	1.90	1.23 a	1.25 a	1.37 ab	1.46 a
4. Dyamin-OSA	0.64	1.17 a	1.23 a	1.16 b	1.32 a
5. Dyamin-OSA	1.28	1.31 a	1.31 a	1.28 ab	1.38 a
6. Agsil 21	0.35	1.16 a	1.26 a	1.40 a	1.41 a
		3 August	17 August	31 August	
1. Control	0	1.35 c	1.15 c	1.22 c	
2. K-silicate	0.95	1.40 bc	1.37 bc	1.42 ab	
3. K-silicate	1.90	1.56 a	1.63 a	1.61 a	
4. Dyamin-OSA	0.64	1.41 bc	1.19 c	1.33 bc	
5. Dyamin-OSA	1.28	1.45 abc	1.23 bc	1.47 ab	
6. Agsil 21	0.35	1.50 ab	1.43 ab	1.51 a	

Means followed by the same letters within the same column are not significantly different at $p = 0.05$.

3.8. Root Growth Characteristics, Biomass, and Viability

All Si treatments increased the root biomass and root volume when compared to the control (Table 8). The application of K-silicate at 0.95 and 1.9 mL m^{-2}, Dyamin-OSA at 0.64 and 1.28 mL m^{-2}, and Agsil 21 at 0.35 mL m^{-2} increased the root biomass by 21.3%, 32.9%, 19.5%, 25.0%, and 28.0%, respectively, when compared to the control.

Table 8. Root growth characteristics, biomass, and viability response to foliar application of three silicon sources in creeping bentgrass putting greens.

Treatment	Rate	Biomass	Length	SA	Diameter	Volume	Viability
	(mL m^{-2})	(mg cm^{-3})	(cm)	(cm^2 cm^{-3})	(mm)	(cm^3 dm^{-3})	(A490 g^{-1} FW)
1. Control	0	1.64 b	90.1 b	5.71 c	0.202 ab	29.1 c	0.51 c
2. K-silicate	0.95	1.99 a	111.7 ab	6.82 bc	0.186 b	32.6 bc	0.68 ab
3. K-silicate	1.90	2.18 a	139.8 a	9.02 a	0.211 ab	48.8 a	0.77 a
4. Dyamin-OSA	0.64	1.96 a	114.0 ab	8.32 ab	0.231 a	48.3 a	0.63 bc
5. Dyamin-OSA	1.28	2.05 a	110.2 ab	7.71 abc	0.230 a	45.9 ab	0.68 abc
6. Agsil 21	0.35	2.10 a	137.5 a	9.12 a	0.214 ab	48.3 a	0.78 ab

Means followed by the same letters within the same column are not significantly different at $p = 0.05$, using Fisher's protected least significant difference test. SA = surface area.

Foliar application of K-silicate at 1.90 mL m^{-2} and Agsil 21 at 0.35 mL m^{-2} also increased the root length and surface area (Table 8). The application of K-silicate at 0.95 and 1.90 mL m^{-2} and Agsil 21 at 0.35 mL m^{-2} improved the root viability by 33.3%, 51.0%, and 52.9%, respectively, when compared to the control.

4. Discussion

The results of this study indicate that foliar application of the Si products enhanced the turf quality relative to the control, especially during dry-down cycles, in creeping bentgrass putting greens. The Si treatments increased the turf quality ratings by 5.8% to 10.1% relative to the control at the end of the 2nd dry-down cycle. This is consistent with previous studies in controlled environments with creeping bentgrass by Schmidt et al. (1999) [16], Merewitz and Liu (2021) [11], and Zhang et al. (2024) [2] and tall fescue (Eneji et al., 2008) [6]. The creeping bentgrass was subjected to continuous heat (usually 35/25 C (day/night)) and drought stress for about 2 months, while it experienced heat and drought for about ten days in each dry-down cycle in field conditions. To our knowledge, this is the first study on Si effects on creeping bentgrass putting greens in field conditions during summer stress in the U.S. transition zone. The quality decline of creeping bentgrass and other cool-season turfgrass species during summer is mainly due to the heat and drought stress in the U.S. transition zone. Si has been used to alleviate the quality decline of creeping bentgrass [2,5]. Previous studies showed that exogenously applied Si to foliage may deposit in cell walls to stabilize cell structure and may also reduce the transpiration of cellular water loss. In addition, Si application may enhance antioxidant capacity, such as SOD activity; suppress ROS toxicity; and alleviate oxidative injury and protect photosynthetic function and pigments [2].

The data of this study show that Si application increased the leaf PE and chlorophyll and carotenoid content when compared to the control. The Si treatments alleviated drought-induced decline in photosynthetic function by protecting photochemical efficiency and chlorophyll as well carotenoids, which function as antioxidants. This is in general agreement with previous studies that indicated that Si application increased the PE and chlorophyll content in drought-stressed creeping bentgrass [2,16] and Kentucky bluegrass [14]. Under abiotic stress, excess energy from sunlight may be targeted to oxygen molecules, thus producing various ROS, which cause damage to cell macromolecules (protein, lipids, and nucleic acids, etc.). Plants could suppress ROS-induced oxidative injury through effective antioxidant defense mechanisms. Plants with greater a antioxidant capacity may exhibit better tolerance to abiotic stress [1]. The results of this study indicate that K-silicate at 1.90 mL m^{-2}, Dyamin-OSA at 1.27 mL m^{-2}, and Agsil at 0.35 mL m^{-2} promoted antioxidant SOD activity when compared to the control. This is consistent with previous studies with creeping bentgrass by Schmidt et al. (1999) [16], Zhang et al. (2024) [2], and Merewitz and Liu (2019) [11] and with Kentucky bluegrass by Bae et al. (2017) [14]. Although antioxidant responses to Si treatments varied in previous studies depending on many factors, it seems that Si application could increase the antioxidant enzyme SOD activity of creeping bentgrass, especially under abiotic stress. SOD is considered the first line of defense against ROS toxicity, and our previous study showed that SOD activity positively responded to Si application [2]; therefore, SOD activity was analyzed as a biomarker for antioxidant enzyme activity in response to Si treatment in this field study. In addition, carotenoids are antioxidant metabolites. The results of this field study support findings from a controlled environment, which showed that Si application increased the carotenoid content, especially during drought stress. In the present study, Si treatments reduced the leaf H_2O_2 level, possibly due to a greater capacity of antioxidant defense systems, including SOD activity. Wang et al. (2021) [23] indicated that Si treatment-induced improvements in drought stress tolerance may be related to the increase in root hydraulic conductance, because suppression of H_2O_2 by a greater antioxidant capacity may increase root hydraulic conductance. Bazem et al. (2002) [24] noted that H_2O_2 is an essential factor in the formation of suberin lamellae, which establishes a hydrophobic barrier in the endodermis and exoderms of roots. Under drought stress, Si treatments reduce H_2O_2 content and suberin lamella formation and thus increase water permeability. The Si-induced decline in ROS production under drought stress corresponded with an increase in antioxidant capacity, especially SOD activity [2,25]. This suggests that Si may improve plant tolerance to drought and heat stress by improving root structure water permeability and by metabolic regulation of antioxidant capacity and photosynthetic activity. Our results in field conditions confirm the findings on the positive effects of Si on antioxidant enzyme activity from previous studies under controlled environments [11,16]. This suggests that Si application may trigger an antioxidant defense system to suppress ROS-induced oxidative injury of plant photosynthetic function and pigment and cell membrane integrity.

Plants can uptake substantial amounts of Si from soil via passive and active pathways. Abiotic stress may reduce root growth and viability as well as available metabolic energy, resulting in a decline in Si uptake and assimilation. The data from this field study show that foliar application of the Si products improved leaf Si concentration when compared to the control, as measured on 3 August, 17 August, and 31 August (Table 6). K-silicate at 1.90 mL m^{-2} and Agsil at 0.35 mL m^{-2} consistently increased the leaf Si content when compared to the control, as measured in August. This is in general agreement with previous studies [2,10,26,27]. Si is immobile in the plant and accumulates in old leaves and does not translocate to new developing ones [5,28]. Exogenously applied Si could be partially taken up by leaf tissues and directly assimilated for the growth of new leaves. This suggests that exogenous Si could improve plant tolerance to drought by strengthening cell walls, enhancing antioxidant defense, and protecting photosynthetic function.

Silicon is absorbed via roots as H_4SiO_4 and is translocated to shoots rapidly either apoplectically or simplistically through root tissues [4]. A large root system with great root viability may effectively absorb Si from soil solutions. The results of this study indicate that all Si treatments increased the root biomass and root volume when compared to the control. K-silicate at 1.90 mL m^{-2} and Agsil 21 at 0.35 mL m^{-2} also increased the root length and surface area and root viability. Since K-silicate contained 4.7% silica and Agsil 21 contained 26.5% silica, the silica rate was 89.3 mg m^{-2} for K-silicate at 1.90 mL m^{-2} and 92.75 mg m^{-2} for Agsil 21. It appears that a silica rate of about 90 mg m^{-2} is effective in inducing root response. In addition, the results of this study show that all three forms of Si (potassium silicate, monosilicic acid from Dyamin-OSA, and silica from Agsil 21) exhibited positive effects on physiological fitness, leaf Si accumulation, and root growth and viability of creeping bentgrass putting greens. This is in general agreement with previous studies by Eneji et al. (2008) [6] with silica gel and potassium silicate and Zhang et al. (2024) [2] with monosilicic acid. There were no reports on the effects of Si on root growth in creeping bentgrass putting greens. A greenhouse pot study with Kentucky bluegrass showed that foliar application of Si applied at 0.1 and 1.0 mM consistently increased root fresh and dry weights under drought stress conditions. This suggests exogenous Si may improve endogenous Si concentrations by enhancing root growth and function, especially under abiotic stress environments. Turfgrass practitioners could incorporate Si in bio-product-based nutrition programs to improve creeping bentgrass putting green quality during summer stress in this transition zone.

5. Conclusions

Foliar application of the three Si products improved turf quality, physiological fitness, and root growth and viability in creeping bentgrass putting greens under field conditions. Among the treatments, K-silicate at 1.90 mL m^{-2}, Dyamin-OSA at 0.64 mL m^{-2}, and Agsil 21 at 0.35 mL m^{-2} tended to have greater beneficial effects than other Si treatments. Exogenous Si may improve drought tolerance by enhancing root growth and viability, Si uptake and assimilation, and antioxidant capacity and by protecting photosynthetic function. The

Si treatment-induced reduction in H_2O_2 may be associated with the improvement of root viability and water permeability under drought stress. The results of this study suggest that foliar application of Si products could be a practical approach to improve creeping bentgrass persistence during the summer months in the U.S. transition zone and other regions with a similar climate. These results with creeping bentgrass in this study are applicable to other cool-season turfgrass species, such as Kentucky bluegrass and perennial ryegrass. Future research should focus on optimum Si rates for different regions with different climates for creeping bentgrass and other cool-season turfgrass and warm-season turfgrass species.

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