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## Research Article

# Effects of Silicate Fertilizer on Growth and Silicic Acid Accumulation in Turfgrass

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## Abstract

**Background and Objective:** Silicon is a globally important soil nutrient to crops for productivity and alleviates the effects of environmental stress in higher plants, including turfgrass. Therefore, this study aimed to evaluate the effects of silicate fertilizer in two turfgrass species, *Zoysia matrella* 'Wakaba' and creeping bentgrass, *Agrostis stolonifera* 'Nightlife', in terms of plant growth, coverage rate, silicic acid content in shoots and roots and root elongation and growth in both turfgrasses. **Materials and Methods:** The *Z. matrella* and creeping bentgrass were cultured under SiO<sub>2</sub> treatments (0, 500 and 1,000 g m<sup>-2</sup>). Plant height, number of shoots, stolon length and coverage were investigated in *Z. matrella*. In addition, dry matter weight and silicic acid content of shoots, stolons and roots were analyzed in *Z. matrella* and the same was done in creeping bentgrass except stolons. **Results:** Silicate treatments tended to increase plant growth, coverage rate, dry matter weight and shoot silicon content in *Z. matrella* and increased silicon content of shoots and roots in creeping bentgrass. **Conclusion:** Higher silicic acid content in the shoots than in roots of *Z. matrella* and creeping bentgrass confirms that they are both silicon-accumulating species. Moreover, the content of silicic acid in *Z. matrella* increased in the order of roots, stolons and shoots which suggests the existence of a gradual silicic acid transport mechanism in *Zoysia* species.

**Key words:** Creeping bentgrass, silicate, silicic acid, silicic acid accumulation, turfgrass, *Zoysia matrella*

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**Competing Interest:** The authors have declared that no competing interest exists.

**Data Availability:** All relevant data are within the paper and its supporting information files.

## INTRODUCTION

Silicon (Si) is the second most abundant element in the earth's crust<sup>1</sup> and is found in the form of silicates ( $\text{SiO}_2$ ) in association with soil minerals<sup>2</sup>. Although there is a high presence of Si in soil, it is only available for entry or uptake into higher plants as silicic acid,  $\text{Si}(\text{OH})_4$ <sup>3</sup>. Silicon is a major constituent in higher plants<sup>4</sup> and is a beneficial substance<sup>5</sup> for plant growth and crop productivity, especially under stressful conditions<sup>6</sup>. Monocot plants are strong accumulators and grasses in particular, have high silicic acid content as they absorb a large amount of silicic acid compared to most plant species<sup>4</sup>. Investigation on the role of Si on higher plants, especially in rice, a major crop in Japan, was conducted by Japanese scientists in the early 20th century<sup>7,8</sup>. Among the highlights, increasing the Si content in rice has helped improve photosynthetic capacity and dry matter production<sup>9</sup>, suppression of rice blast disease<sup>10</sup> and improvement of root elongation and oxidative power<sup>1,4</sup>. In addition, a gene for silicic acid absorption in rice was also found and isolated<sup>11-13</sup>.

There has been a growing attention on the application of silicate fertilizer in turfgrasses<sup>14</sup> for the improvement of turf quality, tolerance to environmental stress and increase pest resistance in important crops. Increasing silicic acid content have improved turfgrass quality in perennial ryegrass, Kentucky bluegrass and tall fescue<sup>5,15,16</sup>, increased treading and cold resistance of seashore paspalum<sup>17,18</sup>, treading resistance of Bermudagrass<sup>14</sup>, disease resistance and environmental tolerance in St. Augustine grass<sup>19</sup> and increased roots of Kentucky bluegrass<sup>20</sup>. The effect of silicate application on creeping bentgrass, a cool-season turfgrass, increased the accumulation of silicon content in foliage and roots<sup>21</sup>, increased coverage and dry matter weight<sup>22,23</sup>, increased roots<sup>22</sup> and improved resistance to dollar spots<sup>24</sup>. On the other hand, silicate application in zoysiagrass, a warm-season turfgrass, increased the accumulation of silicic acid in the shoots<sup>15,25</sup>, improved insect resistance and resistance to large patch disease<sup>14,25</sup>. However, there were no reports on silicic acid accumulation on both shoots and roots.

Therefore, this study examined the effects of silicate application in two turfgrass species, *Zoysia matrella* and creeping bentgrass (*Agrostis stolonifera* L.) in terms of turf growth, coverage, dry matter weight and silicic acid content. In addition, this study investigated the effects of silicic acid on root elongation and growth in both, turfgrasses.

## MATERIALS AND METHODS

**Sample material:** The test materials were *Zoysia matrella* 'Wakaba'<sup>26</sup>, a warm-season turfgrass and creeping bentgrass (*Agrostis stolonifera* L.) 'Nightlife' (Kaneko Seed Co. Ltd.), a cool-season turfgrass species. Wakaba is a variety that was registered in 2015 and is excellent in initial growth, fast coverage rate and stay-green in winter. 'Nightlife' has a deep green leaf color and has an excellent heat resistance (Kaneko Seed Co., Ltd. catalog).

**Cultivation method:** The test was carried out at the Kibana Field Science Center, University of Miyazaki (31°83'N, 131°41'E), Miyazaki Prefecture, Japan in December, 2018–2019. Stolons were removed from fully-grown sods of *Z. matrella* and were washed with running water. Each stolon containing 2 nodes was planted in a 9 cm diameter pot with soil and pre-cultured in a glasshouse. After pre-cultivation, plants in pots were transplanted in cultivation trays (25x25x6 cm) filled with culture soil and mixed with fertilizer (N: P: K = 8: 8: 8) at 30 g m<sup>-2</sup> in March 5, 2019. In addition, culture soil (burnt soil) was supplied at about 1.0 cm from the bottom of the tray. On the same day, 10 g m<sup>-2</sup> of seeds of creeping bentgrass were sown in cultivation tray containing similar culture soil mixture as earlier described. The same chemical fertilizer was supplied to cultivation trays at 3 g m<sup>-2</sup> in 8 May, 2019. In the test area, a silica fertilizer (super energy, Fuji Silysia Chemical Ltd. Japan) containing 90% of  $\text{SiO}_2$  guaranteed components was applied to 0, 500 and 1,000 g m<sup>-2</sup> fertilization treatments with three replications per each treatment. Irrigation was performed twice for 20 min each in the morning and evening using a timer-type automatic sprinkler. Control and aeration were not performed during the test period.

**Investigation:** The investigated traits of *Z. matrella* include plant growth attributes such as plant height, number of shoots, stolon length, coverage rate, dry weight and silicic acid contents of shoots, stolons and roots. On the other hand, creeping bentgrass was investigated with the following traits: plant height, shoot and root dry matter weights and silicic acid contents of shoots and roots. Data gathering was carried out 3 months after both *Z. matrella* and creeping bentgrass were transplanted in cultivation trays. The grass height was measured in 8 June 2019 using a Fujisaki lawn grass height meter (Daito Techno Green Co., Ltd.). The coverage was calculated by extracting the green part from the

photographed image. That is, a RAW data format image was acquired using a digital camera (Panasonic LUMIX GX-1) from a height of 32 cm above the tray's surface with a tray shielded from light to avoid reflection of direct sunlight. The obtained RAW data was developed and trimmed with SILKPIX Developer Studio 3.1SE (Ichikawa Soft Laboratory Co., Ltd.), a high-quality RAW development software. In order to extract the green color element, the image element RGB (red, green, blue) was calculated as the product of the difference between green and red and the difference between green and blue using Adobe Photoshop 7.0 (Adobe Inc.). Green was calculated as white. Binary processing was performed with the color of black as black and calculated as the ratio of white per unit area. The duration of the investigation covered a period of three months (March–June, 2019) and the survey date for coverage rate was from 8 May–8 June, 2019.

To measure dry matter weight, *Z. matrella* samples were fractionated into each plant fractions (shoots, stolons and roots), while for creeping bentgrass, two hole cups (183.1 cm<sup>2</sup>) with a hole cutter were used for sampling to separate grass shoots and roots. All plant fractions were dried at 80°C for 48 hrs in a forced-air oven and the dry matter weight per 100 cm<sup>2</sup> was calculated.

To measure silicic acid content, roots of the samples were thoroughly washed to remove the soil and plant samples were fractionated into each plant fraction for *Z. matrella* (shoots, stolons and roots) and creeping bentgrass (shoots and roots) prior to drying at 80°C for 48 hrs. After drying, each part was

crushed with a mill and mixer (TML161 TESCOM Co., Ltd.) and the crushed sample was wet-decomposed by the sulfuric acid-hydrogen peroxide water method ["Soil, water quality and plant analysis method" p. 251 (Japan Soil Association)]. That is, 1.0 g of a crushed sample, 8 mL of sulfuric acid and 2 mL of distilled water were added to a conical beaker and while heating at 140–160°C on a hot plate, hydrogen peroxide solution was appropriately added to decompose until transparent and then filtered using a filter paper [No. 6 φ125 mm (ADVANTEC, Toyo Roshi Kaisha, Ltd.)]. After the filtration, the filter paper with the remaining residue was put into a crucible and burned in a burner and a magnetic muffle furnace and then burned in an electric furnace [FP32 (Yamato Scientific Co., Ltd.)] set at 600°C for 2 hrs. The weight of the obtained residue was measured and the ash content was calculated as the silicic acid content.

**Meteorological data:** The meteorological data at the test site in Miyazaki City, Miyazaki Prefecture in 2019 (Fig. 1) was obtained from the Japan Meteorological Agency website. The normal year is the average of the 29 years observed climate conditions from 1981–2010 at the station. Temperatures remained high during all test periods except mid-April and early May, especially in late-April which recorded 1.7°C higher than the average. On the other hand, precipitation in 2019 remained low except in early March, late-April and mid-May, where precipitation was three times higher than the average rainfall. Rain commenced at the beginning of the experiment

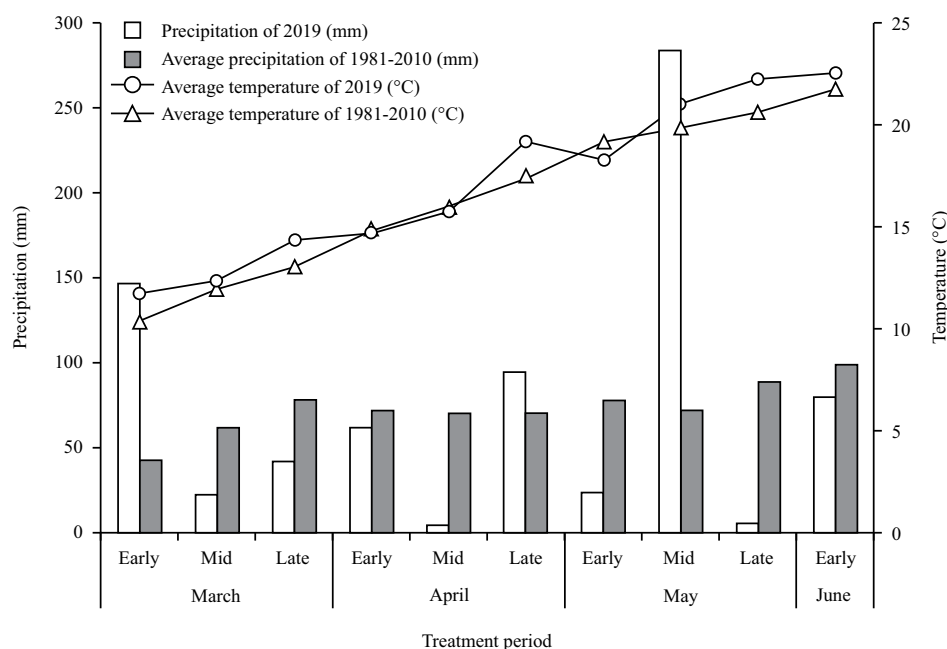


Fig. 1: Temperature and precipitation in Miyazaki City during the treatment period (March–June, 2019)

in early-March which have allowed germination and early plant growth to proceed smoothly. On the other hand, precipitation in mid-April and late-May was about 1/15th than that of the average rainfall but this problem was avoided since automatic irrigation was used during this stage of the experiment. Although the temperature and precipitation during the test period were different from the normal value, the test could be performed without affecting the growth of turfgrass.

## RESULTS

Table 1 summarizes the effects of silicate treatment on plant height, number of grass blades and stolon length of *Z. matrella* 'Wakaba'. Plant height, number of grass blades and length of stolons of *Z. matrella* tended to increase as the amount of silicate treatments increased. In particular, plants under 1,000 g m<sup>-2</sup> silicate treatment showed significantly ( $p < 0.05$ ) higher values for all traits than the untreated plants. The plant height was approximately 1.7 times higher ( $9.73 \pm 0.65$  cm) for the 1,000 g m<sup>-2</sup> silicate-treated plants than that of untreated plants ( $5.73 \pm 1.55$  cm) and the number of grass blades was approximately 2.5 times for the

1,000 g m<sup>-2</sup> silicate-treated plants than that of the non-treated plants ( $252 \pm 82$  and  $631 \pm 23$  cm). The stolon length was  $747 \pm 59$  cm for the 1,000 g m<sup>-2</sup> silicate-treated plants and  $265 \pm 98$  cm for untreated plants. Only the number of grass blades significantly increased in the 500 g m<sup>-2</sup> silicate-treated plants compared to the untreated plants.

Figure 2 summarizes the effect of silicate treatment on the coverage rate of *Z. matrella* 'Wakaba'. Although the effect of silicate treatment was not observed 1 month after transplantation, the covering rate of all treated and untreated plants tended to increase in 2–3 months after transplantation. Plant coverage rate of *Z. matrella* was significantly ( $p < 0.05$ ) higher at 38.6% coverage rate for the 1,000 g m<sup>-2</sup> silicate treatment and about 2.6 times greater than the untreated plants at 14.6% coverage rate in June, 2019. Although *Z. matrella* treated with 500 g m<sup>-2</sup> silicate tended to show a higher coverage rate than the untreated plants, their difference was not significant. The images show the increase in coverage of *Z. matrella* fertilized with 500g m<sup>-2</sup> and 1,000g m<sup>-2</sup> silicate from 1–3 months after transplantation in Fig. 3. This confirms that the degree of increase in coverage of treated plants is high.

Table 1: Effect of silicic acid treatment on plant height, number of shoots and stolon length of *Zoysia matrella*

Treatment (g m <sup>-2</sup> )	Plant height (cm)	Number of shoots	Stolon length (cm)
0	$5.73 \pm 1.55^a$	$252 \pm 82^a$	$265 \pm 98^a$
500	$7.80 \pm 1.41^{ab}$	$537 \pm 77^b$	$558 \pm 141^{ab}$
1,000	$9.73 \pm 0.65^b$	$631 \pm 23^b$	$747 \pm 59^b$

\* Values within the columns followed by the same letter(s) are not significantly different at  $p < 0.05$  by Tukey HSD test

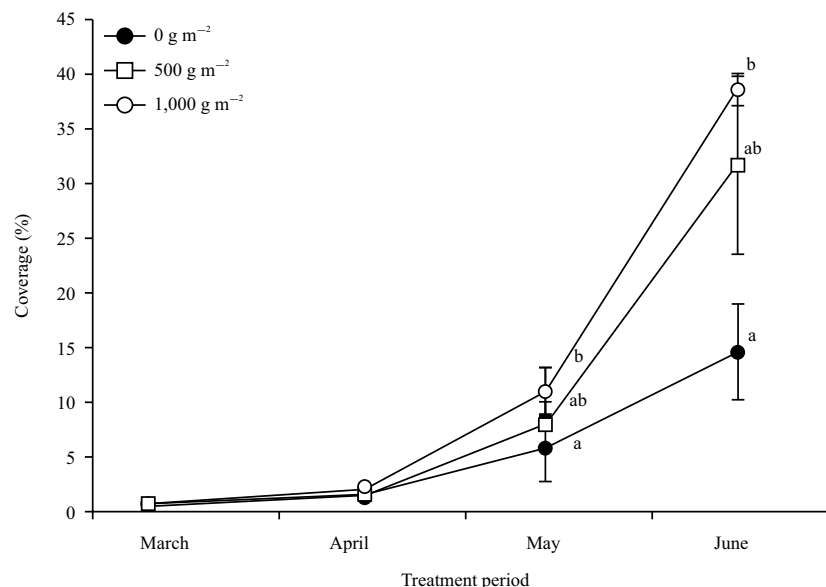


Fig. 2: Effect of silicic acid treatments on the coverage rate of *Zoysia matrella*

\*Line points with the same letter(s) are not significantly different at  $p < 0.05$  by Tukey HSD test



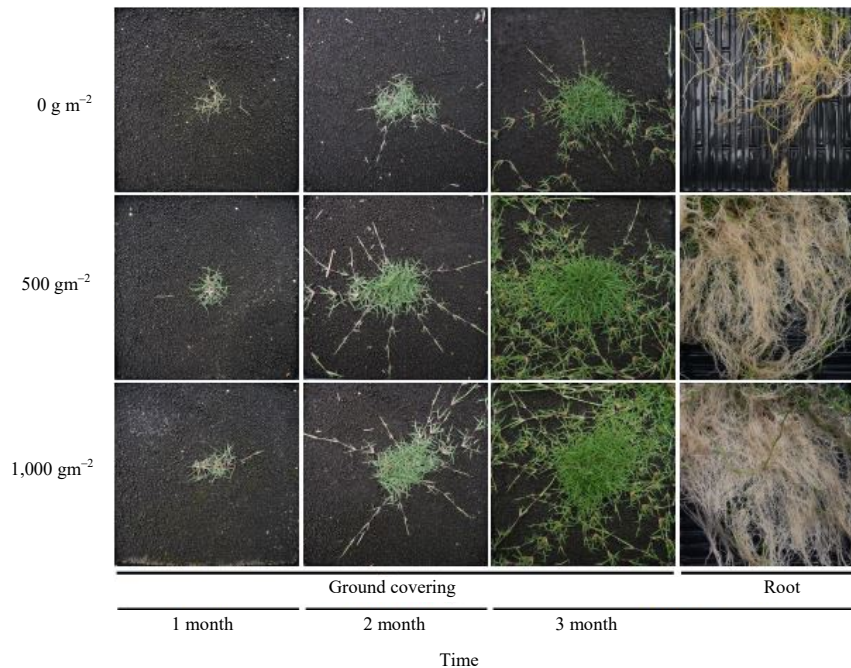


Fig. 3: Comparison of 1–3 months ground covering and 3-months-old root after planting in each silicic acid treatment in *Zoysia matrella*

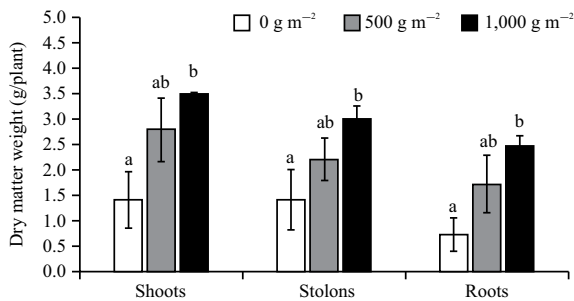


Fig. 4: Effect of silicic acid treatment on dry matter weight of erect stems, stolons and roots of *Zoysia matrella*

\*Bars with the same letter(s) are not significantly different at  $p < 0.05$  by Tukey HSD test

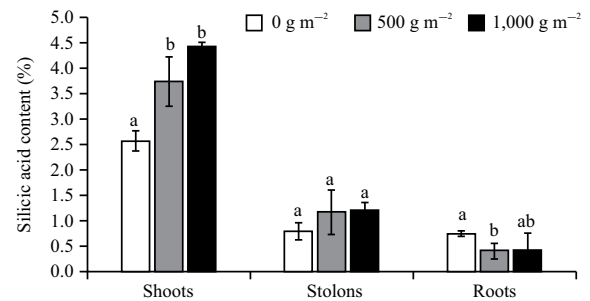


Fig. 5: Effect of silicic acid treatment on silicic acid content of shoots, stolons, and roots of *Zoysia matrella*

\*Bars with the same letter(s) are not significantly different at  $p < 0.05$  by Tukey HSD test

Figure 4 summarizes the effect of silicate treatments on dry weight of shoots, stolons and roots of *Z. matrella* 'Wakaba'. The dry weight of all plant fractions significantly increased in the 1,000 g m<sup>-2</sup> silicate-treated plants as compared with the untreated plants. Treatment with 500 g m<sup>-2</sup> of silicate showed an intermediate value between plants treated with 1,000 and 0 g m<sup>-2</sup> and it was observed that the dry weight tended to increase as the amount of treatment increased. It can be confirmed from the root images (Fig. 3) that root mass increased in both the silicate-treated plants.

Silicate fertilizer treatments affected the silicic acid content of shoots, stolons and roots of *Z. matrella* 'Wakaba' (Fig. 5). The silicic acid content in shoots significantly increased ( $p < 0.05$ ) with increasing silicate treatments at 500 and 1,000 g m<sup>-2</sup> and the silicic acid content rate was about 1.7 times that of the untreated plants. On the other hand, the content of silicic acid in the roots significantly ( $p < 0.05$ ) decreased in the treated plants but there was no significant difference in the content in the stolons. The content of silicic acid in *Z. matrella* was higher in the order of roots, stolons and shoots.

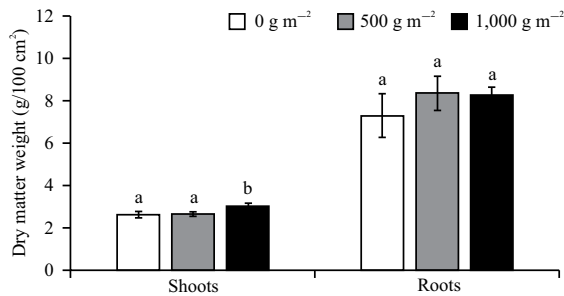


Fig. 6: Effect of silicic acid treatment on dry matter weight in shoots and roots of creeping bentgrass

\*Bars with the same letter(s) are not significantly different between characters at  $p < 0.05$  by Tukey HSD test

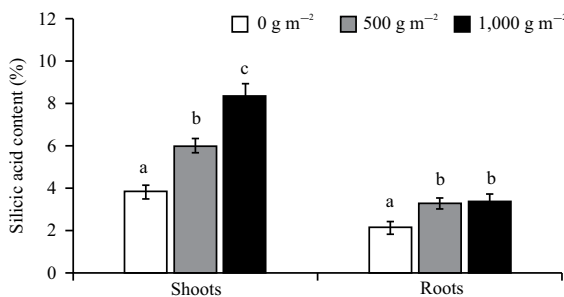


Fig. 7: Effect of silicic acid treatments on the content of silicic acid in shoots and roots of creeping bentgrass

\*Bars with the same letter(s) are not significantly different between characters at  $p < 0.05$  by Tukey HSD test

Figure 6 show that silicate treatment affected the shoot and root dry weights of creeping bentgrass (*Agrostis stolonifera* 'Nightlife'). The dry matter weights of shoots and roots of creeping bentgrass were significantly higher in plants treated with  $1,000 \text{ g m}^{-2}$  silicate than with untreated ( $0 \text{ g m}^{-2}$ ) plants. However, no significant differences were found between root dry matter weights of creeping bentgrass under 500 and  $1,000 \text{ g m}^{-2}$  silicate treatments.

Figure 7 summarizes the effect of silicate treatment on the content of silicic acid in the shoots and roots of creeping bentgrass. The silicic acid content in the shoots significantly increased after the application of 500 and  $1,000 \text{ g m}^{-2}$  silicate. On the other hand, the silicic acid content of shoot in plants treated with  $1,000 \text{ g m}^{-2}$  silicate was significantly more than double than that of the untreated ( $0 \text{ g m}^{-2}$ ) plants. In addition, the silicic acid content of roots significantly increased in both plants treated with 500 and  $1,000 \text{ g m}^{-2}$  silicate as compared with the untreated ( $0 \text{ g m}^{-2}$ ) plants. Moreover, the silicic acid content was higher in the shoots than in the roots in all 3 treatments. However, there was no significant difference in terms of grass height of creeping bentgrass in all treatment intervals (Table 2).

Table 2: Effect of silicic acid treatment on plant height of creeping bentgrass

Treatment ( $\text{g m}^{-2}$ )	Plant height (cm)
0	$5.97 \pm 0.12$ ns
500	$5.73 \pm 0.12$ ns
1,000	$5.87 \pm 0.39$ ns

ns: No significant

## DISCUSSION

Generally, the addition of silicate fertilizer can increase silicic acid content and has been reported to have a growth effects in various plants by restoring foliar damage and improving growth in food crops such as rice, cucumber and tomato<sup>27</sup>. In this study, improvement in covering rate and the growth of shoots, stems and roots were observed after silicate application in *Z. matrella* 'Wakaba'. It has also improved turfgrass growth<sup>14,17,25</sup> and improved turf quality<sup>22</sup> as exemplified by the improvement in fraying resistance, insect resistance and disease resistance in the *Z. japonica* cultivar 'Miyako'. Turfgrass is heavily stressed on a daily basis because it is managed by regular cutting. The fact that the application of silica fertilizer showed improvement in the growth, and increased the coverage of the erect stems and the stolons suggest that it is highly resistant to such stress. On the other hand, silica fertilizer improved disease resistance in other turfgrasses by reducing gray leaf spots in St. Augustine grass, Bipolaris leaf spots in bermudagrass<sup>14</sup> and brown patches in creeping bentgrass<sup>28</sup>, indicating that the ability to increase tolerance to various environmental stressors can be achieved by supplementing turfgrass with silicates. In the future, a detailed study on the effects of silicate treatment in *Z. matrella* 'Wakaba' in terms of environmental resistance such as treading resistance and disease resistance will be conducted.

Plants are divided into three types based on their silicic acid absorption properties. Silicon accumulator plant, such as grasses, silicon non-accumulator plant, such as tomato and an intermediate type plant, such as cucumber<sup>1</sup>. Silicic acid accumulation in gramineous plants and ferns are positively absorbed using energy and since the mechanism exists in the roots, the silicic acid content of roots is lower than that of foliage<sup>29</sup>. The content of silicic acid in *Z. matrella* and creeping bentgrass in this study was higher in the shoots than in the roots, suggesting that these two grass species are also silicon-accumulating plants. It has been reported that silicic acid content increased by adding silicate fertilizers in many grass species<sup>15,16,22,25</sup>. However, there are no reports on the effects of silicic acid accumulation on the shoot and root parts of *Z. matrella* and creeping bentgrass. This study investigated the silicic acid content in the stems and roots as

well as in the stolons, which are the specific turf organs in *Z. matrella*. As a result, it was found that the silicic acid content in not only the roots but also the stolons was lower than that in the shoots, and the content of silicic acid of *Z. matrella* gradually increased in the order of root, stolons and shoots. Interestingly, the content of silicic acid in roots of treated plants was significantly lower than that of the untreated plants, suggesting that zoysiagrass may have a specific silicate transport mechanism.

In the present study, the application of silicate increased the dry weight and coverage of shoots, stolons and roots in *Z. matrella*, especially at 1,000 g m<sup>-2</sup>. The results were similar to those reported in many turfgrasses such as creeping bentgrass<sup>22,23</sup>, St. Augustine grass<sup>14,19</sup>, Kentucky bluegrass<sup>16,20</sup>. However, silicate application did not affect the dry weight of *Z. japonica* 'Miyako'<sup>15</sup> and is considered to be related to the lower fertilizer application rates at 100 and 300 g m<sup>-2</sup> silicate reported by Saigusa *et al.*<sup>25</sup> as compared to the addition amounts of 500 and 1,000 g m<sup>-2</sup> in this study. On the other hand, although dry weight of *Z. matrella* plants treated with 500 g m<sup>-2</sup> silicate fertilizer was higher than that of untreated plants, no significant difference was observed between the two treatments. Therefore, it is necessary to apply 500 g m<sup>-2</sup> or more of silicate to increase the dry weight of *Z. matrella*. On the other hand, the shoot dry weight of creeping bentgrass in this study was significantly high at 1,000 g m<sup>-2</sup> silicate treatment, which is similar to that reported by Kim and Kim<sup>22</sup> and Kim *et al.*<sup>23</sup>. Previous reports on creeping bentgrass<sup>22,23</sup> and other turfgrasses<sup>16,20</sup> confirmed that root elongation and growth were enhanced by the application of silica fertilizer. Although no significant difference in the root dry matter weight of creeping bentgrass was found in this study, it tended to increase in other reports. However, results varied because the previously reported test<sup>23</sup> was conducted on already established grass or sod grass such as in a golf course, whereas the creeping bentgrass in this study was cultivated at the initial growth stage after sowing.

## CONCLUSION

In this study, it was found that *Z. matrella* and creeping bentgrass had higher silicic acid content in the shoots than in the roots, indicating that they are silicic acid-accumulating plants, similar to rice. Silicate fertilizer was useful as growth promoting material in both zoysiagrass and creeping bentgrass and it promoted root elongation and foliage growth in both species and improved coverage of zoysiagrass.

## SIGNIFICANCE STATEMENT

This study discovered that both *Zoysia matrella* and creeping bentgrass (*Agrostis stolonifera*) are both silicon-accumulator species. The silicic acid content of *Z. matrella* gradually increased in the order of roots, stolons and shoots and the content of silicic acid in roots of treated plants was significantly lower than that of the untreated plants. This suggests that zoysiagrass may have a specific silicate transport mechanism.

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