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## Evaluating Pre-silicon Treatment to Alleviate Drought Stress and Increases Antioxidative Activity in *Zoysia japonica*

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**ABSTRACT.** This study was performed to determine the effects of silicon on zoysiagrass after the application of drought stress. The daily amount of water or silicon solution was 150 ml per a pot. For 14 days, plants were treated with 0.1 and 1.0 mM silicon (Si) and with distilled water for control and the drought only-treatment. Afterward, the plants in Si and drought treatment were exposed to a 21-day under drought stress condition but the plants in control received water. The results indicated that the growth and the moisture and chlorophyll contents decreased in the drought only-treatment and 0.1 mM Si compared to the control. However, 1.0 mM Si showed an increase in the growth with a significant increase of water and chlorophyll contents. The MDA and H<sub>2</sub>O<sub>2</sub> concentrations and electrolyte leakage decreased, while the radical scavenging capacity increased in 1.0 mM Si. 1.0 mM Si showed little to no differences in the growth and no differences in water and chlorophyll contents, electrolyte leakage, MDA and H<sub>2</sub>O<sub>2</sub> concentrations and antioxidant capacity compared to the control. These results suggested that application of silicon is useful for drought tolerance improvement of zoysiagrass under drought that is occurring in turf fields.

**Key words:** Drought stress, Electrolyte Leakage, Hydrogen Peroxide, Malondialdehyde, Zoysiagrass

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### Introduction

Turfgrass is a unique group of plant species cultivated for ornamental decoration around us for recreation in golf courses and sports fields, and for land coverage and protection (Chen et al., 2009). Turfgrass has a relatively excellent adaptability to various environments, and the use of turfgrass has been extended in terms of range and areas (Bea et al., 2013). The main warm-season turfgrass species belonging to zoysiagrasses (*Zoysia* spp.) are native to the East and South East, and they are naturally widespread in Korea (Engelke et al., 1983). Research on drought stress on turfgrass relating to water saving is needed because of rapid desertification and occurrence of droughts, due to the changes in climate from global warming. Turfgrass water management is important for maintaining high turfgrass quality under conditions of water scarcity.

Dryness, which is one of the major causes of environmental stress, inhibits photosynthesis in plants and thereby reduces growth (Iturbe-Ormaetxe et al., 1998), while a decline in the

carbon assimilation rate is known to induce oxidative stress (Bartosz, 1997; Holmberg and Bulow, 1998) by promoting the production of reactive oxygen species (ROS) and by increasing the flow of photosynthetic electrons from O<sub>2</sub> (Asada, 1994). Harmful ROS produced in cells inhibit the electron transport activity of chloroplasts and mitochondria. It also changes in the content and composition of chlorophylls and damage to the photosynthetic apparatus suppress the photochemical activity, while a decline in the enzymatic activity in the Calvin cycle reduces the photosynthetic activity of plants (Monakhova and Chernyad, 2002). Also, lipid peroxidation destroys cell membranes as well as the antioxidant defense system that responds to oxidative damage such as changes in cell composition and proteins, and oxidative stress inhibits growth (Iturbe-Ormaetxe et al., 1998; Fu and Huang, 2001).

Silicon existing in the Earth's crust is classified as the most abundant element following oxygen (Epstein, 1994). Although silicon is not considered as an essential element for plant growth, a number of studies have reported it as an important factor in plants and proved that it plays an important role in

the resistance mechanisms of plants against environmental stress (Epstein, 1999; Savant et al., 1999). Silicon was also designated as an element with a beneficial influence on plant growth and development and crop yield (Liang, 1999; Epstein, 1994) and required in large amounts for Gramineae and crops, in particular (Lewin and Reimann, 1969; Parry and Smithson 1964). The benefits of silicon include disease control (Raid et al., 1992), mitigation of heavy metal toxicity (Neumann and Nieden, 2001), and alleviation of salt stress (Zhu et al., 2004; Liang et al., 2003).

When silicon was added to rice (*Oryza sativa* L.) under drought stress, the transpiration rate and membrane permeability were reduced to prevent loss of water from the leaves (Agarie et al., 1998). Also, a comparison of dry wheat (*Triticum aestivum* L.) treated and untreated with silicon showed that the wheat treated with silicon maintained better moisture content and had an increased dry matter content that helped increase resistance to drying (Gong et al., 2003).

Silicon is reported to mitigate oxidative damage to functional and antioxidant defense molecules and maintain physiological processes such as photosynthesis (Gong et al., 2005). Treating soybeans (*Glycine max* L.) that were placed under drought stress with silicon resulted in an increase in the fluidity of stomata and photosynthetic activity, and in turn enhanced their growth (Shen et al., 2010).

There are still an insufficient number of studies on drought stress on grass, and there are only few studies done on the effect of silicon on the drought resistance of grass. Thus, this study was conducted to determine the effect of silicon on the drought stress of zoysiagrass.

## Materials and Methods

### Plant materials and treatments

This study was conducted from May 2011 to November 2011. The plant used in this study was zoysiagrass (*Zoysia japonica* Steud.) Twelve stolons cut into 3-node length each were planted in the sand filled plastic pot with a diameter of 12 cm and a height of 10 cm. The river sand had been washed for a week with distilled water before using and 680 g of sand were filled into a plastic pot. The temperature in the plant growth chamber was maintained at 25°C day and night, RH 60~80%, light conditions at 12 hours and light intensity at 390  $\mu\text{mol m}^{-2} \text{s}^{-1}$ . The plants were watered once a day from the head with distilled water of 150 ml  $\text{pot}^{-1}$  and were fertigated twice a week with Hoagland nutrient solution (Hoagland, 1950). The composition of the Hoagland's concentrate was 236 g  $\text{L}^{-1}$  1 M  $\text{Ca}(\text{NO}_3)_2 \cdot 4\text{H}_2\text{O}$ , 101 g  $\text{L}^{-1}$  1 M  $\text{KNO}_3$ , 136 g  $\text{L}^{-1}$  1 M  $\text{KH}_2\text{PO}_4$ , 246 g  $\text{L}^{-1}$  1 M  $\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$ , 18.4 g  $\text{L}^{-1}$  0.05 M Na EDTA, 87 g  $\text{L}^{-1}$  0.5 M  $\text{K}_2\text{SO}_4$ , 1.72 g  $\text{L}^{-1}$  0.01 M  $\text{CaSO}_4 \cdot 2\text{H}_2\text{O}$ , 2.86 g  $\text{L}^{-1}$   $\text{H}_3\text{BO}_3$ , 1.81 g  $\text{L}^{-1}$   $\text{MnCl}_2 \cdot 4\text{H}_2\text{O}$ , 0.22 g  $\text{L}^{-1}$   $\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$ , 0.08 g  $\text{L}^{-1}$   $\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$ , 0.02 g  $\text{L}^{-1}$   $\text{H}_2\text{MoO}_4 \cdot 4\text{H}_2\text{O}$  (85%

Molybdic acid). At 120 days after planting with approximately 80% of coverage rate of the stolons, 0.1 and 1 mM of Si (DSi) had been treated with 150 ml  $\text{pot}^{-1}$  once a day for 14 days with  $\text{Na}_2\text{SiO}_3$  as the source of Si but plants of control (CT) and drought (DR) treatment had been received only distilled water. The pH of the solution was adjusted to 5.5 using  $\text{H}_2\text{SO}_4$ . Afterward, for 21 days, drought treatment was performed but control received 150 ml of distilled water once a day. Measurements were done right after the 21-day drought treatment. The treatments were done in completely randomized design with 6 replications and, for statistical analysis, Duncan Multiple Range Test was performed using SAS (v.9.1, Cary, NC, USA) to analyze the differences between the treatments.

### Measurement of plant growth and the water content

The shoot and root length and fresh and dry weight of shoot, stolon, and rhizome were measured to determine the effect of silicon treatment on drought stress. The shoot and root were dried for 48 hours at 80°C and weighed.

In order to determine the water content, the fresh weights (FW) of shoot and root were measured. The samples were dried for 48 hours at 80°C and weighed to determine the dry weight (DW). The water content was calculated by substituting the following equation:

$$\text{Water content (\%)} = (\text{FW}-\text{DW}) / \text{FW} \times 100$$

### Analysis of the chlorophyll contents

To determine the chlorophyll contents, 0.05 g of the fresh samples were homogenized using 100% acetone solvent in accordance with Lichtenthaler's (1987) method and centrifuged for 10 minutes at 4°C and 2,800 xg. Using spectrophotometer (UV-1800, Shimadzu, Japan), the absorbance of the supernatant was measured at wavelengths of 661.6, 644.8 and 470 nm, and the concentrations of chlorophyll and carotenoid were calculated by using the following equation:

$$\text{Chl a} = (11.24 \times A_{661.6}) - (2.04 \times A_{644.8})$$

$$\text{Chl b} = (20.13 \times A_{644.8}) - (4.19 \times A_{661.6})$$

$$\text{Chl a+b} = (7.05 \times A_{661.6}) + (18.09 \times A_{644.8})$$

### Analysis of electrolyte leakage and malondialdehyde and hydrogen peroxide concentrations

As for the electrolyte leakage, 0.1 g of the fresh sample was immersed in 20 ml of distilled water and the electrical conductivity was measured (Electrolyte Leakage,  $\text{EL}_0$ ). Then, it was shaken at 120 rpm for 24 hours using an agitator and extracted before the electrical conductivity was measured again ( $\text{EL}_{24}$ ). It was then extracted for 24 hours in an 80°C water bath and its electrical conductivity was measured ( $\text{EL}_t$ ). The degree of electrolyte leakage was calculated by substituting the

following equation:

$$EL (\%) = (EL_{24} - EL_0) / (EL_t - EL_0) \times 100$$

The oxidation of lipid was measured from the concentration of malondialdehyde (MDA), which is a decomposition product of unsaturated fatty acids. As for the MDA concentration, 0.2 g of the fresh sample was homogenized in 5 ml of 5% trichloroacetic acid solvent and centrifuged (12,000 xg) at 4°C for 20 minutes in accordance with the method used by Heath and Pacher (1968). 2 ml of 0.6% thiobarbituric acid solvent was added to 2 ml of supernatant, which was then boiled in an 80°C water bath for 15 minutes. The extracted solvent was then centrifuged for 10 minutes at 12,000 xg and 4°C before measuring the absorbance at wavelengths of 450, 532, 600 nm using the spectrophotometer to be substituted into the following equation.

$$MDA (\text{nmol L}^{-1}) = 6.45 (A_{532} - A_{600}) - 0.56 A_{450}$$

As for the concentration of hydrogen peroxide (H<sub>2</sub>O<sub>2</sub>), 0.3 g of the fresh sample was homogenized in 0.1% trichloroacetic acid (TCA) solvent and centrifuged for 15 minutes at 12,000 xg and 4°C in accordance with the method used by Velikova et al (2000). 0.5 ml of 10 mM potassium phosphate buffer (pH 7.0) and 1.0 ml of 1.0 mM KI were added to 0.5 mL of supernatant before measuring the absorbance at a wavelength of 390 nm using the spectrophotometer. The hydrogen peroxide concentration was calculated from the standard curve.

#### Measuring of DPPH free radical scavenging activity

The Electron Donating Ability (EDA) test using DPPH ( $\alpha, \alpha$ -diphenyl- $\beta$ -picrylhydrazyl, Sigma-Aldrich chemical, Co. USA) was performed on the extract to determine its antioxidant capacity. To be specific, 16 mg DPPH reagent was dissolved in 100 ml of absolute ethanol to prepare  $4 \times 10^4$  M DPPH solution. Next, 3 ml of ethanol was added to the extract

with the leaf sample prepared in 0.2 ml and the solution was mixed with 0.8 ml DPPH reagent before being powerfully shaken for 10 seconds and left at room temperature for 10 minutes. The absorbance was measured at a wavelength of 525 nm using a spectrophotometer and the difference between the absorbance of the group containing the sample and that of the group without the sample was indicated as a percentage (%) to measure the EDA. The calculation method is as follows:

$$EDA (\%) = [1 - ABS / ABC] \times 100$$

(ABS: absorbance of the group containing sample; ABC: absorbance of the group without sample)

## Result and Discussion

### Effect of silicon on plant growth, the water and chlorophyll contents under drought stress

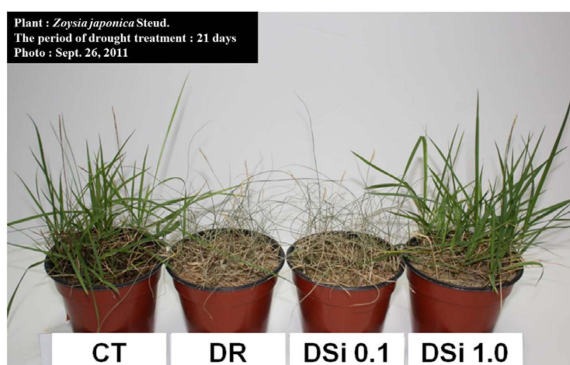
Table 1 shows the effect of silicon treatment on zoysiagrass that was placed under drought stress. Compared to the control (CT), the plants exposed to a 21-day drought condition (DR) showed a significant decrease in the shoot and root lengths and the fresh and dry weight of shoot, stolon and root. When turfgrass was treated with 0.1 and 1.0 mM silicon (DSi) before drought stress, DSi 0.1 showed the downward tendencies of the growth as DR, whereas DSi 1.0 showed an increase in shoot and root length by 20.9% and 12.9%, respectively. In addition, with respect to the shoot and root of zoysiagrass treated by DSi 1.0, the fresh weight increased by 170.3% and 45.5% and the dry weight by 29.0% and 33.3%, respectively. No significance was found between DR and DSi 1.0 on the fresh and dry weight of stolon. DSi 1.0 showed similar tendencies as CT in terms of growth. From this, it was thought that silicon treatment mitigates the growth inhibition caused by drought stress. After a 21-drought treatment, plants treated by DR and DSi 0.1 withered completely whereas those in DSi 1.0 showed healthy and greeny condition as control plants (Fig. 1). Compared to CT, the water content of shoot, rhizome and

**Table 1.** The effect of silicon treatment on the growth of zoysiagrass (*Zoysia japonica*) under drought.

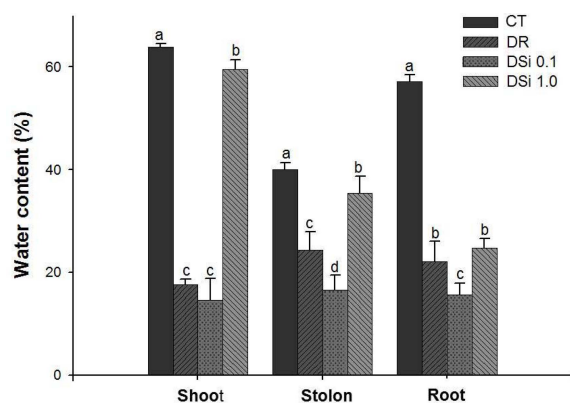
Treatment <sup>z</sup>	Shoot length (cm)	Root length (cm)	Shoot (g 10 plants <sup>-1</sup> )		Stolon (g 78.5 cm <sup>-2</sup> )		Root (g 10 plants <sup>-1</sup> )	
			Fresh weight	Dry weight	Fresh weight	Dry weight	Fresh weight	Dry weight
CT	18.0a <sup>y</sup>	13.4a	1.05a	0.38a	0.30a	0.18a	0.32a	0.14a
DR	14.8c	9.3d	0.37b	0.31b	0.23ab	0.17a	0.11c	0.09b
DSi 0.1	15.4c	10.0c	0.42b	0.35a	0.24bc	0.20a	0.16b	0.13a
DSi 1.0	17.9a	10.5b	1.00a	0.40a	0.28ab	0.18a	0.16b	0.12a

<sup>z</sup>Si of 0.1 and 1 mM (DSi) treated once a day with 150 ml pot<sup>-1</sup> for 14 days but plants of control (CT) and drought (DR) treatment treated with distilled water only. Afterward, for 21 days, drought treatment was performed but control received 150 ml of distilled water once a day. Measurements were done right after the 21-day drought treatment.

<sup>y</sup>Mean separation within columns by Duncan's multiple range test,  $P=0.05$ .

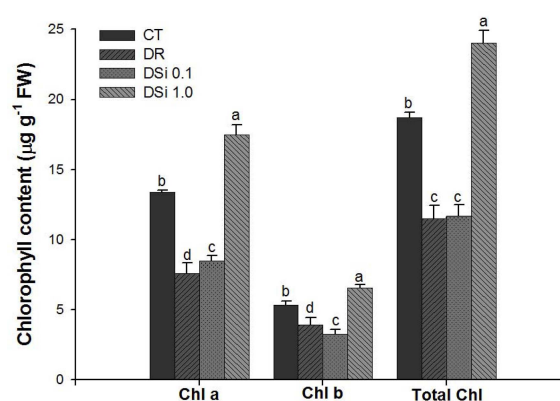


**Fig. 1.** Effect of silicon on the drought resistance of zoysiagrass (*Zoysia japonica*) after a 21-day of drought. Si of 0.1 and 1 mM (DSi) treated once a day with 150 ml pot<sup>-1</sup> for 14 days but plants of control (CT) and drought (DR) treatment treated with distilled water only. Afterward, a 21-day drought treatment was performed but control received 150 ml of distilled water once a day.



**Fig. 2.** Effect of silicon on the water content of each part of zoysiagrass (*Zoysia japonica*) under drought. Si of 0.1 and 1 mM (DSi) treated once a day with 150 ml pot<sup>-1</sup> for 14 days but plants of control (CT) and drought (DR) treatment treated with distilled water only. Afterward, a 21-day drought treatment was performed but control received 150 ml of distilled water once a day. Bars indicate standard error of the mean. Dissimilar letters indicate mean separation within columns by Duncan's multiple range test at  $P=0.05$ .

root of DR decreased significantly (Fig. 2). Plant growth is controlled by cell division, growth rate, and supply of organic and inorganic compounds needed to synthesize new protoplasm and cell walls (Kramer and Boyer, 1995). Cell growth is especially related to turgor pressure, while stem and leaf growth is suppressed by lack of water (Salisbury and Ross, 1992). Reduced water content inhibits photosynthesis and decreases the respiration rate and other metabolic rates related to enzymes. In other words, reduced water content is accompanied by loss of turgor pressure, cessation of cell growth, stoma obstruction, decreased photosynthetic activity and decrease in other basic metabolic activities. When zoysiagrass was treated with 0.1 and 1.0 mM silicon before being dried, DSi 0.1 had reduced water content compared to DR, whereas the water content of the shoot and rhizome of



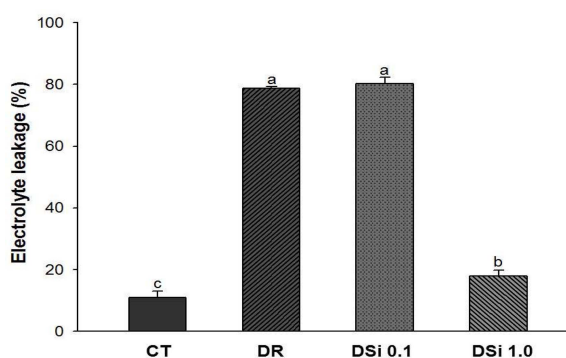
**Fig. 3.** Effect of silicon on the chlorophyll content of zoysiagrass (*Zoysia japonica*) under drought. Si of 0.1 and 1 mM (DSi) treated once a day with 150 ml pot<sup>-1</sup> for 14 days but plants of control (CT) and drought (DR) treatment treated with distilled water only. Afterward, a 21-day drought treatment was performed but control received 150 ml of distilled water once a day. Bars indicate standard error of the mean. Dissimilar letters indicate mean separation within columns by Duncan's multiple range test at  $P=0.05$ .

DSi 1.0 increased by 138.1% and 45.5%, respectively, with no significant difference in the water content of the root compared to DR. The water content of the shoot, rhizome and root of DSi 1.0 decreased by 6.7%, 11.5% and 72.5%, respectively, compared to CT. Based on the biggest decrease in the root, it was determined that the water loss in the shoot is lower than the root. This can be explained by the fact that silicon is absorbed by plant root as silicic acid  $[\text{Si}(\text{OH})_4]$ , a non-polar monomer molecular, at under pH 9 (Ma and Takahashi, 2002). After being taken up by the root, silicon is immediately transported to the shoot along with the transpiration current and accumulates in the leaf cells (Yosida, 1965). Because silicon mainly remains in the outer wall of the epidermal cells in the leaves, it helped reduce the water loss caused by transpiration in the stomata (Agarie et al., 1998; Savant et al., 1999). Compared to CT, the total chlorophyll content decreased significantly in DR (Fig. 3). Photosynthesis is especially sensitive to water content (Iturbe-Ormaetxe et al., 1998). When there is an insufficient amount of moisture due to drying, the stomata become closed and chloroplasts are not supplied with carbon dioxide from the atmosphere (Agarie et al., 1998; Savant et al., 1999). This in turn leads to a decrease in the carbon assimilation rate and production of the harmful ROSs in cells that inhibit electron transfers in chloroplasts and mitochondria, and the changes in the chlorophyll content and damage to the photosynthetic apparatus result in reduced photosynthetic capacity of the plant (Anjum et al., 2011; Iturbe-Ormaetxe et al., 1998; Miyashita et al., 2005). When turfgrass was treated with 0.1 and 1.0 mM silicon before being dried, DSi 0.1 showed similar tendencies as DR, whereas the total chlorophyll content in DSi 1.0 increased by 113.2% compared to DR and by 27.4% compared to CT. It was shown

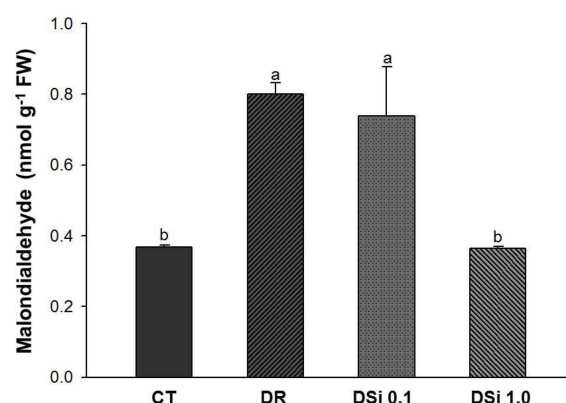
that the higher the silicon content, the higher the chlorophyll content. According to Gong et al. (2005), the improvement of photosynthesis of wheat plants applied with silicon was associated with non-stomatal factors, the increase in activities of photosynthetic enzymes and ribulosebiphosphate carboxylase, as well as the chlorophyll content under drought stress. Shen et al. (2010) also reported that treating soybean with silicon after applying stress increases the fluidity of the stomata and the photosynthetic activity and this may be due to the correlation between silicon and the increased photosynthetic enzyme activity and the chlorophyll and anthocyanin contents in stress conditions. Adatia and Besford (1986) reported that adding silicon to cucumber grown in a nutrient recirculation system caused an increase in the chlorophyll content and the activity of ribulosebiphosphate carboxylase.

#### Effect of silicon on electrolyte leakage, the malondialdehyde and hydrogen peroxide concentrations under drought stress

When zoysiagrass was treated with 0.1 and 1.0 mM silicon before being dried, electrolyte leakage of DSi 0.1 increased greatly with almost no similarity to DR (Fig. 4). Electrolyte leakage of DSi 1.0 was lower compared to DR and slightly higher compared to CT. When plant is placed under stress such as drought stress, reactive oxygen lead to oxidative damage similar to lipid peroxidation (Scandalios, 1993). Unsaturated fatty acids that make up cell membranes get especially severely damaged by  $^1\text{O}_2$  (singlet oxygen) or  $\cdot\text{OH}$  (hydroxyl radical) that get generated during the oxidation process, and lipid hydroperoxides are produced from cellular structures. This causes a decrease in the fluidity of the cell membrane and promotes electrolyte leakage from inside the



**Fig. 4.** Effect of silicon on the electrolyte leakage of zoysiagrass (*Zoysia japonica*) under drought. Si of 0.1 and 1 mM (DSi) treated once a day with 150 ml pot<sup>-1</sup> for 14 days but plants of control (CT) and drought (DR) treatment treated with distilled water only. Afterward, a 21-day drought treatment was performed but control received 150 ml of distilled water once a day. Bars indicate standard error of the mean. Dissimilar letters indicate mean separation within columns by Duncan's multiple range test at  $P=0.05$ .

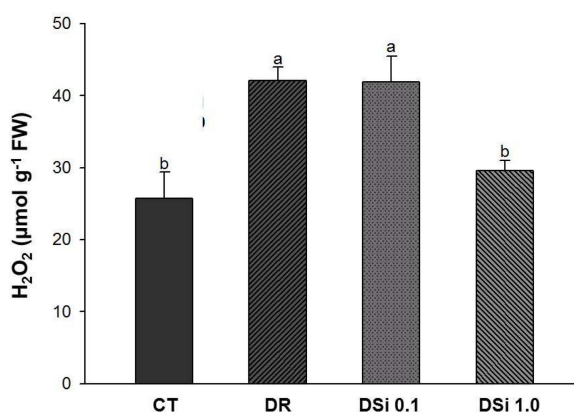


**Fig. 5.** Effect of silicon on the MDA concentration of zoysiagrass (*Zoysia japonica*) under drought. Si of 0.1 and 1 mM (DSi) treated once a day with 150 ml pot<sup>-1</sup> for 14 days but plants of control (CT) and drought (DR) treatment treated with distilled water only. Afterward, a 21-day drought treatment was performed but control received 150 ml of distilled water once a day. Bars indicate standard error of the mean. Dissimilar letters indicate mean separation within columns by Duncan's multiple range test at  $P=0.05$ .

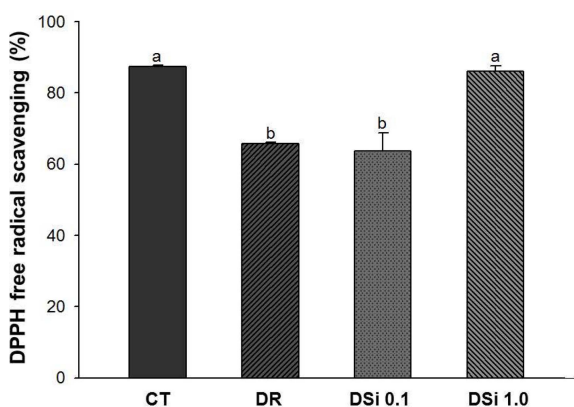
cells to bring a secondary damage to the membrane proteins (Moller et al., 2007). In contrast, treating turfgrass with silicon prior to drying decreased the electrolyte leakage. Shen et al. (2010) proved that treating soybeans with silicon after a stress decreased electrolyte leakage, while Zhujun et al. (2004) proved that treating cucumber with silicon after salt stress resulted in reduced permeability of the plasma membrane and lipid peroxidation of the cell membrane as well as facilitation of growth by helping to maintain the original function and prevent changes of the cell membrane. When zoysiagrass was treated with 0.1 and 1.0 mM silicon before being dried, DSi 0.1 showed no significant differences from DR, whereas the MDA concentration in DSi 1.0 decreased compared to DR with no difference from CT (Fig. 5).

MDA is the final product of the process, in which ROS generated by oxidative stress causes lipid peroxidation of the cell membrane, and is an indicator that shows the degree of oxidation of membrane lipids (Halliwell and Gutteridge, 1989; Scandalios, 1993). Zhang et al. (2005) reported that the lipid peroxidation was increased by oxidative stress, which in turn reduced the quality of Kentucky Bluegrass, and high temperature and dryness can cause a simultaneous increase of electrolyte leakage and lipid oxidation in plant leaves (Liu et al., 2008). The results also showed that drying can lead to severe electrolyte leakage and significant increase in the MDA concentration, from which it was determined that changes in the cell membrane are one of the physiological reactions to drought stress. When turfgrass was treated with 1.0 mM silicon before being dried, the MDA concentration decreased. Agarie et al. (1998) reported that treating crops in the rice family with silicon after drought and heat stress prevented the





**Fig. 6.** Effect of silicon on the H<sub>2</sub>O<sub>2</sub> concentration in zoysiagrass (*Zoysia japonica*) under drought. Si of 0.1 and 1 mM (DSi) treated once a day with 150 ml pot<sup>-1</sup> for 14 days but plants of control (CT) and drought (DR) treatment treated with distilled water only. Afterward, a 21-day drought treatment was performed but control received 150 ml of distilled water once a day. Bars indicate standard error of the mean. Dissimilar letters indicate mean separation within columns by Duncan's multiple range test at  $P=0.05$ .



**Fig. 7.** Effect of silicon on DPPH free radical scavenging activity of zoysiagrass (*Zoysia japonica*) under drought. Si of 0.1 and 1 mM (DSi) treated once a day with 150 ml pot<sup>-1</sup> for 14 days but plants of control (CT) and drought (DR) treatment treated with distilled water only. Afterward, a 21-day drought treatment was performed but control received 150 ml of distilled water once a day. Bars indicate standard error of the mean. Dissimilar letters indicate mean separation within columns by Duncan's multiple range test at  $P=0.05$ .

reduced function and changes to the cell membrane to enhance the structural stability of membrane lipids.

Compared to CT, the H<sub>2</sub>O<sub>2</sub> concentration significantly increased in DR (Fig. 6). When zoysiagrass was treated with 0.1 and 1.0 mM silicon before being dried, DSi 0.1 showed no significant differences from DR, whereas the H<sub>2</sub>O<sub>2</sub> concentration decreased significantly compared to DR with almost no differences in the concentration compared to CT. According to Davis (1995), oxidative stress on plants leads to the production of harmful ROSs such as superoxide anion ( $\cdot O_2$ ), hydrogen peroxide (H<sub>2</sub>O<sub>2</sub>), hydroxyl radical ( $\cdot OH$ ) and

singlet oxygen ( $^1O_2$ ), which cause oxidative damage. An excessive accumulation of ROS in plant cells lead to the oxidation of molecular substances such as DNA, protein and lipid (Fadzilla et al., 1997; Liang et al., 2003). The results showed that the H<sub>2</sub>O<sub>2</sub> concentration increased in turfgrass after drought stress and decreased after silicon treatment. This was consistent with the findings of Gong et al. (2005), who reported that the H<sub>2</sub>O<sub>2</sub> concentration increased significantly in dried wheat, while it decreased significantly after silicon treatment. Oxidative stress from drying led to an increase of H<sub>2</sub>O<sub>2</sub>, which is an ROS, and likely caused physiological and visible disturbances in turfgrass. Such disturbances are thought to be mitigated by silicon. Compared to CT, the oxygen radical scavenging activity of DR was significantly lower (Fig. 7).

#### Effect of silicon on DPPH free radical scavenging activity under drought stress

When zoysiagrass was treated with 0.1 and 1.0 mM silicon before being dried, DSi 0.1 showed no significant differences from DR, whereas DPPH free radical scavenging of DSi 1.0 was significantly higher than DR with no significant differences from CT. Rios et al. (2008) reported that an increase in DPPH free radical scavenging is a defense mechanism employed in addition to an increase of antioxidant substances to eliminate ROS generated due to oxidative stress, and this is used to determine the antioxidant capacity of plants. Treating turfgrass with silicon led to an increase of the free radical scavenging activity, which was similar to the findings of Liang et al. (2003), who reported that treating barley with silicon after applying an oxidative stress such as salt stress led to reduced lipid peroxidation and increased antioxidant enzyme activity in the roots to eliminate ROS.

From this study, it was found that silicon significantly reduced the growth and water content of zoysiagrass and this reduction may attribute to increased drought-induced oxidative stress which resulted in an increase in membrane permeability and the MDA and hydrogen peroxide concentrations. These physiological interference and visually noticeable disturbances in zoysiagrass can be mitigated by the addition of silicon. Thus, we suggested that application of silicon is useful for drought tolerance improvement of zoysiagrass under drought that is occurring in turf fields.

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