Influence of Humic Substances on Irrigation Frequency and Phosphate Absorption of Creeping Bentgrass Putting Greens

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Abstract

Humic substances (HS) reportedly enhance moisture retention of soil. However, no information is available as to the effects of HS on sand-based golf course putting greens. HS have acidic functional groups (COOH) that are reaction sites on the molecule. An greenhouse experiment was conducted to evaluate the effect that the acidic functional groups in humic acid, tannic acid and citric acid have on the volumetric water content of sand, and the phosphorus (P) content of ‘Dominant’ creeping bentgrass (Agrostis palustris L.). Bentgrass plugs were grown in calcareous sand and irrigated with 250 mg·L⁻¹ C solutions of each of the organic acid products. Irrigation occurred when the volumetric water content of the soil reached 5%. Soil moisture was measured with a HydroSense® water content sensor for 3 months. Phosphate was added as KH₂PO₄ at 50 kg·ha⁻¹ 2 months into the experiment. The irrigation interval was longer between watering for humic acid, but rarely differed between the treatments. Daily soil moisture percentage following irrigation with all organic acid treatments was different from the control, with humic acid retaining moisture longer on average. Humic acid, tannic acid, and citric acid increased tissue concentrations of P, with tannic acid having the highest percent increase. Further research is necessary to evaluate the residual effects the organic acids may have on longevity in the soil.
Introduction

Creeping bentgrass (*Agrostis palustris* L.) is used on golf course putting greens in the Intermountain West region of the United States. The climate and calcareous soil of this region can impose difficult growing conditions on this cool-season turfgrass species. The summer months of June through August have high temperatures, low humidity and little rainfall that intensify drought conditions. These climactic factors contribute to the amount of supplemental irrigation needed by creeping bentgrass.

The transpiration gradient created by these climactic factors can cause wilting of leaf tissue and the loss of root mass during periods of high drought stress. Although water is needed in large amounts by bentgrass to meet evaporative demands, turfgrass managers are required to conserve water. Water shortages have been created by climatic and hydrological conditions that are worsened by increasing public consumption. Therefore, golf course managers attempt to use as little water as possible, but still maintain extremely high quality turf demanded by golfers. Creeping bentgrass putting greens require frequent nutrient applications in order to be maintained at a high level.

Fertilization can be dictated by the media of the putting green root-zone. Calcareous sand used in the Intermountain West can create nutrient deficiencies for creeping bentgrass. High amounts of calcium carbonate buffer soil pH levels above 7.2 (Grossl and Inskeep, 1992), rendering phosphorus and some micronutrients less available. Nutrient deficiencies reduce plant health, and can hinder drought avoidance mechanisms. One method that may reduce irrigation and provide nutrient availability for bentgrass during stress is the application of organic products such as humic acid.
Humic acid is a fraction of organic matter categorized as humic substances. Humic substances are divided into fractions of humic acid, fulvic acid, and humin. Humic substances are long-chained molecules whose shape is dependant on pH (Stevenson, 1982). Humic acid is soluble at pH>2, fulvic acid is soluble at all pH values and humin is insoluble at all pH values. Humic substances are a natural heterogeneous substance that is characterized as being yellow or black in color, of high molecular weight, and refractory (Aiken et al., 1985).

The application of humic substances on creeping bentgrass has shown positive growth responses. A bentgrass putting green must continue growing and producing carbohydrates during times of stress. It has been shown that applying humic acid to creeping bentgrass resulted in increased photosynthesis (Liu et al., 1998; Liu and Cooper, 2000; Nardi et al., 2002). By continuing to produce carbohydrates bentgrass may be more adept at drought avoidance. One way a plant can avoid drought stress is with an extensive root system.

Increased rooting can extract more water and nutrients from the soil. In a study using foliar applied humic acid in a sand media, the root mass increased 45% and root depth 15% (Cooper et al., 1998). Also, root enzyme activity was increased in creeping bentgrass, suggesting that root respiration was influenced by humic acid (Liu et al., 1998; Liu and Cooper, 2000). On a putting green with low nutrient retention, an increase in root structure might obtain more nutrients. Nutrients must be in solution for absorption by roots, and humic substances can keep nutrients in solution longer. Humic and fulvic acids can form complexes with phosphate making inorganic phosphorus more available.
(Zalba and Peinemann, 2002). Humic acid can also coat soil particles preventing sorption of phosphate onto constituent surfaces (Grossl and Inskeep, 1992).

A tissue analysis would measure the availability of nutrients with the application of humic substances. There was no increase of P with humic acid applied to hydroponic creeping bentgrass plugs (Liu et al., 1998), but P tissue concentration did increase in sand by applying it to, or incorporating it into the soil (Cooper et al., 1998; Liu and Cooper, 2000). Overall, applications of humic substances provide growth responses that may be beneficial to creeping bentgrass in times of drought.

Growth responses and improved plant heath are important, but can humic substances reduce the amount of supplemental irrigation needed by creeping bentgrass during the summer? No studies have been done to evaluate this claim, and research can determine if humic acid has an effect on moisture retention of creeping bentgrass putting greens. The object of this experiment was to determine if functional group content influenced water holding capacity. The effect humic acid has on phosphate uptake in calcareous sand will also be studied.

**Hypothesis one:** Humic acid increases time between watering of sand putting greens.

**Hypothesis two:** Humic acid increases the rate of phosphorus uptake in calcareous sand.

**Materials and Methods**

The study was conducted in a greenhouse located at Utah State University in Logan, UT. Creeping bentgrass plugs were cut from a research green on 18 July, 2005. The 8-cm-diameter plugs were cut to a depth of 10 cm, and transferred into 1-gal pots
(Figure 1). Calcareous sand (Staker-Parson Companies; Brigham City, UT) was mixed with sphagnum peat moss to match the media of the research green. The media was 90% sand and 10% peat on a volume basis with a pH of 7.8. The plugs were watered with a hose during the establishment period of 5 days.

Three organic acids (humic, tannic, and citric) were applied to the turf as irrigation and evaluated against a control. Deionized water was used to irrigate the control pot. The experiment was completely randomized and was replicated three times. The organic acids were selected based on oxygen functional group content of each molecule (humic acid < tannic acid < citric acid). Of the three, tannic acid is chemically most similar to fulvic acid and was used because of cost restraints. Functional groups are acidic reaction sites, and can be calculated on a molecular basis. Humic acid is 9% functional groups, tannic acid 13% and citric acid 50% per molecule (Grossl and Inskeep, 1992).

To determine if functional group content influenced water holding capacity, the treatment applications had to be equaled. The products used were normalized based on amount of carbon in each molecule. The products included 3.64 g of leonardite humic acid (55% carbon), 3.70 g of galotannic acid (54% carbon) and 5.88 g of citric acid (34% carbon). Each product was dissolved in 2 L of deionized water. This made a 2000 mg·L⁻¹ C stock solution. Deionized water was used to prevent precipitation of solids in the solution. A 1 L solution of 125 mL from the stock solution made a 250 mg·L⁻¹ C watering solution. The watering solution provided adequate amounts of the C treatments based on previous research in the literature. New watering solutions had to be made every three weeks once the study began due to the irrigation frequency of the pots.
An equal irrigation amount was needed for each pot. It was determined that 700 ml was a sufficient volume to saturate the profile. This provided enough drainage to percolate out the bottom of the pot. Drainage was important to prevent buildup of salts in the root zone, and also simulated how a golf course putting green would be flushed during an irrigation event. The total organic carbon content of the soil was not measured during the study.

**Volumetric Water Content.** Soil moisture was measured each morning between 0800 and 1100 HR, seven days a week. A HydroSense® water content sensor (http://www.campbellsci.com/cs620) was used to record data as a percentage of the volumetric water content \((\theta_v)\) (Figure 2). Soil moisture was recorded to a depth of 12 cm for 3 months beginning 22 July, 2005 and ending 31 October, 2005. Field capacity (25%) and permanent wilting point (2.5%) of the sand was not determined, but assumed based on a previous calibration of the sensor. Irrigation occurred independently for each pot. A depletion threshold was set at 5% plant available water. When soil moisture reached 5% according to the sensor, the watering solution was applied. Irrigation treatments began on 22 July, 2005 by saturating each pot with its respective watering solution.

**Phosphorus uptake.** To determine the availability of phosphorus, leaf tissue samples were collected on 28 October, 2005. Because of the small area of each plug, the clippings from each replication were combined into one sample for analysis. A potassium phosphate \((\text{KH}_2\text{PO}_4)\) was applied at 50 kg·ha\(^{-1}\) once beginning 7 October, 2005. Based on the area of each plug, 100 ml was added two or three days after the
previous irrigation to the soil around each pot. This was to prevent leaching of phosphate out of the root zone, and prevented adsorption to plant tissue.

Results

Making the humic acid stock solution required an additional step that the others did not. A small amount of KOH was used to dissolve the 3.64 g of leonardite humic acid crystals before adding it to water. Beginning 9 August, 2005 turf receiving the humic acid had leaf scorching. Turf quality was poor and all three treatments ended up with severe necrosis. The watering solution was analyzed, and the pH was 11.9 due to the KOH. It was discovered that dissolving the leonardite humic acid crystals could be done with deionized water. A new stock solution had to be made but the pH was still 8.4. This was corrected with an addition of 3 ml of 1 M HCl to bring the pH down to 7.8. The tannic acid and citric acid solutions were tested and both had a pH of 7.8. Three new bentgrass plugs had to be used in place of the old humic acid treatments. All the pots in the study were flushed with water on 9 August, 2005. The plugs were inserted and the study resumed after re-randomizing the experiment.

Sand top-dressing material was applied 6 September, 2005 and brushed in by hand to fill in holes created by the sensor probes used every day. Because the turf could not be irrigated directly after this process, the leaf tissue was damaged. The turf was also severely nutrient deficient. For the sake of measuring phosphorus uptake the bentgrass was deprived of nutrients after its establishment. A complete greens fertilizer (18-9-18) was applied at 44 kg·ha⁻¹ of nitrogen on 18 July, 2005, the day the plugs were transplanted. An additional application of urea at a rate of 44 kg·ha⁻¹ of nitrogen was
added on 7 October, 2005. Because the bentgrass was deficient in nitrogen this added to the top-dressing injury. Although turf quality was poor, it began to decline late in the study. Sufficient data was recorded for evaluation purposes.

**Volumetric Water Content.** An average interval between irrigations was examined (Figure 3). The humic acid treatment had the longest irrigation interval of 5.7 days. Citric acid was irrigated every 5.5 days, tannic acid 5.4 days and the control 5.4 days. Only the humic acid was statistically different from the control. However, there were no observable differences between the C treatments. The irrigation intervals were evaluated with a statistical software, but there was no significant differences (p>0.14); (SAS Institute, 1985). Humic acid having the longest interval between irrigations suggested that functional group content may not influence moisture retention.

Daily soil moisture between irrigations was also examined (Figure 4). 1 day following irrigation the humic acid and tannic acid had the highest soil moisture (17.5%), and the control had the lowest (16.2%). Citric acid was the highest on day 2 (14.2%), and humic acid on day 3 (12.0%). On day 4 humic acid (9.4%) and tannic acid (9.5%) had similar moisture levels, with the control (8.8%) the lowest.

**Phosphorus uptake.** Tissue concentration of phosphorus was higher in all the C treatments compared to the control (Figure 5). Tannic acid application resulted in the highest tissue concentration (0.19%). Humic acid and citric acid had equal concentrations (0.17%) and the control was the lowest (0.13%). Because of the poor turf quality, and that this sample was not replicated does not provide statistical evidence for these differences. Although not statistically different the results did provide an interesting trend that shows the potential for humic substances to increase availability of
phosphorus. Additionally the iron concentration in plant tissue also increased for all the C treatments relative to the control. This suggests that humic substances may increase iron availability as well.

**Discussion**

This study provided some interesting trends that will require further research. The next step will be to design a more controlled experiment. Better methods and more measurements may determine if the differences observed from this study have any significance. This study showed that humic acid may retain moisture longer in creeping bentgrass putting greens. Because the largest differences were between the humic acid and the control, functional group content may not influence volumetric water content. Humic acid and citric acid had similar daily soil moisture percentages following irrigation, but humic acid retained water longer on average.

All C treatments had higher plant tissue levels of phosphorus compared to the control. Tannic acid had the highest percentage of phosphorus in leaf tissue, and citric acid was the lowest. High functional group content of citric acid did not influence phosphorus uptake. The longevity of organic materials in the soil was not taken into account in this study for any of the materials. Humic acid persists longer in the soil than citric acid. Future studies should evaluate the total organic carbon content of the media throughout the experiment.

Despite some problems with the methodology, there are interesting trends that were apparent. Top-dressing material should be applied independently for each treatment preceding an irrigation event to prevent tissue damage. Fertilization should be done with
frequent, moderate applications the same way a putting green would be managed. The trends observed suggest that humic acid has the potential to reduce water usage and enhance phosphorus availability to creeping bentgrass putting greens. Further research including field trials will be necessary to support this study’s result.
Literature Cited


Figure 1. Experiment design with *Agrostis palustris* L. creeping bentgrass plugs in one gallon pots.

Figure 2. A HydroSense® sensor used to measure the volumetric water content with 12 cm probes.
Figure 3. Average interval in days between irrigation events of the organic acid treatments.

Figure 4. Average soil moisture percentage for days following an irrigation event.
Figure 5. Amount of phosphorus in plant tissue.