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Progress Report to Georgia Golf Environmental Foundation

Title of Project: Evaluating the Impact of Biological Products on Turf Quality and Soil Biological Health

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1. Background

Due to the need for aesthetics, the maintenance of golf courses entails extensive use of various inputs (e.g., fertilizers, pesticides, wetting agents, plant growth regulators, water). This makes it among the most expensive sector in agriculture (cost of input/acre). Reducing inputs is therefore important for the future of the golf course industry. This is particularly applicable to low and medium size clubs that have limited financial capacity. Reducing input is also important from the point of view of reducing the environmental footprint of golf courses. Environmental concerns have led to the proliferation of biological products that are collectively called biostimulants. These products contain microorganisms (bugs in a jug) and/or organic products that are often marketed as being more sustainable and cheaper alternatives to current products that are commonly used in the golf course industry. This assumes that the biological products are better in stimulating the indigenous soil microorganisms that provide beneficial services. However, there is lack of research in evaluating how effective biological products are and how they affect the health of the turfgrass system and turf quality.

According to United State Department of Agriculture, soil health is defined as, "how well the soil performs its function now and how well the functions are preserved for future use." The way golf courses are managed can result in three scenarios in relation to soil health: it can improve it; it can sustain it; It can degrade it over time. Soil health is evaluated by measuring indicators that are reflective of changes in physical, chemical and biological properties of the soil in response to management practices. Microorganisms are central to the biological health of the soil. Biological soil health indicators are reflective of how well microorganisms perform their function (i.e., how well they provide all the beneficial services mentioned above) in the system. For instance, soil respiration and enzyme activity measurements are indicative of the microorganisms' role in degrading organic matter and recycling nutrients. The goal of this proposal is, therefore, to examine the impact of biological products on turf quality and soil health. We are interested in examining the relationship between soil health and turf quality, i.e., will improvement in soil health lead to improvement in turf quality?

One important consideration when evaluating biological products is the method of application. Products are commonly surface applied, leading to exposure of microorganisms contained in biological products to extreme climatic fluctuations (e.g., heat and UV exposure from sun). This exposure can reduce the survival and establishment of microbial inoculants in the soil. This can

be minimized through subsurface application of the product. One way of achieving this is by using a unique tool such as Air_2G_2 as a delivery system directly to the root zone, which was originally designed to aerate the soil by blasting air below surface but has been modified to inject products. As part of this project, we have started examining if applying the products at the surface vs below surface with Air_2G_2 will make any difference in the performance of the products.

2. Objectives

- Determine the impact of two biological products on turf quality and soil health
- Determine how the performance of the biological products is affected by method of application (surface vs subsurface)

3. Study Approach

3.1. Field Plots and Treatments

Field plots were established on greens in June 2018 at two separate locations – UGA Griffin Campus (A1-A4 Bentgrass) and Rivermont Golf Club (Tifway Bermudagrass) in Johns Creek, GA. Each plot is 8 x 8 ft in dimension. There are seven treatments, with two biological products (see below). The treatments were designed to apply the two biological products both above and below surface with A2G2 injection system. The surface applications of the products were done coupled with and without A2G2 aerification. The biological products were applied on the top of the standard turf management inputs, including fertilization, irrigation and top dressing. The Control treatment received all standard inputs except for the biological products. Each treatment was replicated four times in a randomized complete block design.

The two biological products that are being tested are KaPreRemeD8-NSL (BP1) and KaPreRemeD8-NSP (BP2) from Performance Nutrition (LidoChem, Inc., Hazlet, NJ). They were applied based on the recommendation rates on the labels at 1 and 19 ounces per 1000 ft^2, respectively. KaPreRemeD8-NSP is described as a proprietary mixture containing *Saccharomyces cerevisiae* where as KaPreRemeD8-NSL is described as a proprietary mixture including fulvic acid.

Treatments were applied monthly since June 2018. Plots in Johns Creek have received fourteen treatments so far. Plots at the UGA Griffin campus received only three treatments (June to August 2018), and treatments were stopped thereafter because the grass was not doing well. New plots were then established at Echelon Golf Club in Milton, GA (A1 Bentgrass) in May 2019. Plots at Echelon have received six treatments to date.

The treatments included the following:

1. None-treated control (water) – No product or aerification
2. BP1 surface application without aerification
3. BP2 surface application without aerification
4. BP1 surface application and Air_2G_2 aerification
5. BP2 surface application and Air_2G_2 aerification
6. BP1 subsurface application with Air_2G_2
7. BP2 subsurface application with Air_2G_2

3.2. Sample Analysis

Before the start of the experiment, the biological products and the soils at both locations were tested for some basic properties (e.g., nutrient contents and organic matter) at the UGA's Environmental and Agriculture Services Laboratory (http://aesl.ces.uga.edu). Once treatment application started, samples were collected periodically (early and after multiple applications) from the top 4 inches. The plots were also fitted with automatic soil moisture and temperature sensors (Campbell, Logan, UT).

Turf Quality: Turf quality was assessed by taking images of the plots with a digital camera and analyzing the images with the Assess 2.0 image analysis software (American Phytopathological Society) as percent green cover (ratio of green to total pixels). It provides an objective assessment of the overall turf quality and quantitative data for robust statistical analysis.

Indicators of Soil Biological Health: We monitored biological soil health indicators that are reflective of the activity and abundance of soil microorganisms. The activity indicators include soil respiration (generic indicator of microbial activity) and enzymes that mediate nitrogen and phosphorous transformations (urease and phosphatase). Enzyme activities were measured based on standard protocols (Wallestein and Weintraub, 2008; Tabatabai, 1994). To quantify microbial abundance, DNA were extracted from all the samples with DNeasy PowerSoil kit (QIAGEN, Germantown, MD, USA). Quantitative polymerase chain reaction was employed to quantify the abundance of ammonia-oxidizing bacteria and archaea that are often used as indicators of soil biological health. Increase in indicators is generally considered to be an improvement in soil biological health.

3.3. Disease suppression (new)

An inoculation study was initiated in July 2019 at both locations to test ability of the products to suppress diseases. The top corner of each plot was inoculated with the pathogens that cause dollar spot (*Clarireedia homoeocarpa*) and leaf spot (*Bipolaris sorokiniana*) in Echelon and Rivermont, respectively. The initial inoculation at Rivermont resulted in no leaf spot appearing on any plots, mainly due to high temperatures and lack of moisture. In Echelon, dollar spot appeared on all of the plots, and there no difference among the treatments. Plots were inoculated again at the end of September and beginning of October. We are currently continuing to monitor and rate the spread of disease at both locations as it has to be done multiple times. We expect to share the findings of this section in our final report.

3.4. Statistical analysis

The data were summarized into descriptive statistics (e.g., mean and standard errors). Analysis of variance was carried out to test the statistical significance of the effects of the biological products on turf quality and indicators of soil health at $\alpha = 0.05$.

4. Results Summary

A. Rivermont Golf Club

- Turf quality: The treatments did not significantly impact turf quality as expressed in percent green cover (Fig 1A), meaning the treatment did not improve or negatively impact the turf quality regardless of how they were applied and when they were applied compared to the Control.
- Soil respiration: No significant treatment effect was observed on soil respiration as well, meaning the microbial activity was not positively or negatively affected by any of the treatments (Figure 1B).

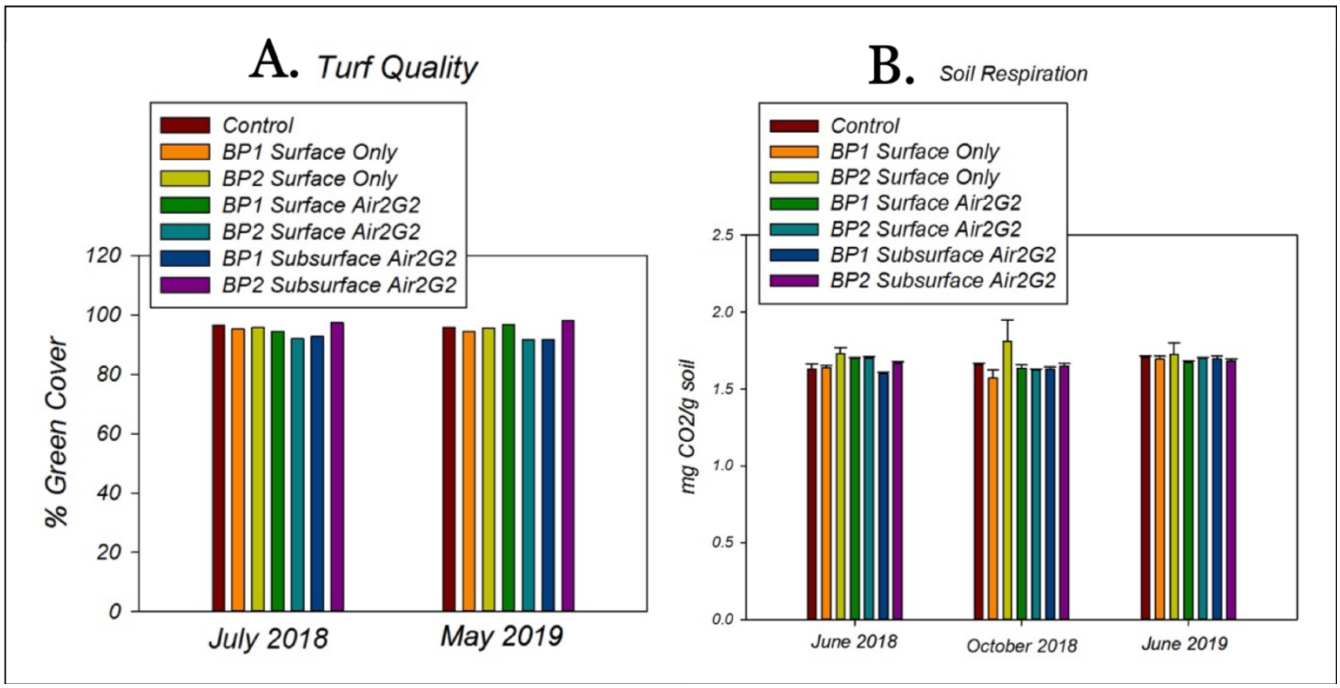

Figure 1: Turf quality (A) and soil respiration (B) in Tifway Bermuda grass in Johns Creek, GA.

- There was no significant treatment effect on phosphatase activity (Table 1). The only significant difference was between sampling times, with phosphatase activity decreasing from June 2018 to June 2019.
- Similar to the phosphatase activity, no significant treatment effect was seen on urease activity as compared to the Control (Table 1). However, there was a significant increase in urease activity from June 2018 to June 2019.
- We also quantified the abundance of ammonia-oxidizing bacteria (AOB) and ammonia-oxidizing archaea (AOA), microorganisms that are commonly used as indicators of biological health, and we did not find any significant treatment effect (data not shown)

Table 1: Phosphatase and urease activities (mean ± 1SE) in Tifway Bermuda turfgrass in Johns Creek, GA. Numbers with same letter suffix are not significantly different from each other. Comparison is valid with in a column.

Treatment	Mean phosphatase activity (µmol P evolved g^{-1} h^{-1})			Mean urease activity (µmol NH_3 evolved g^{-1} h^{-1})		
	June 2018	October 2018	June 2019	June 2018	October 2018	June 2019
NTC	1.318a	0.679a	0.732b	6.85b	10.4b	15.5a
BP1 Surface	0.910a	0.948a	0.820b	5.95b	8.4b	11a
BP2 Surface	1.178a	1.020a	0.980b	8.06b	8.9b	12a
BP1 surface w/ A2G2	1.017a	1.268a	0.678b	8.76b	8.9b	14.5a
BP2 surface w/ A2G2	0.996a	1.59a	0.604b	6.81b	7.4b	11a
BP1 Sub A2G2	0.690a	0.819a	0.626b	7.45b	8.4b	17a
BP2 Sub A2G2	0.816a	1.152a	0.818b	7.38b	8.9b	12.5a

B. Echelon Golf Club

- Turf quality: There was no significant treatment effect on turf quality or soil respiration in the field plots in Milton, GA (Figure 1), i.e., the treatment did not improve or negatively impact the turf quality or microbial activity regardless of how they were applied and when they were compared to the control.

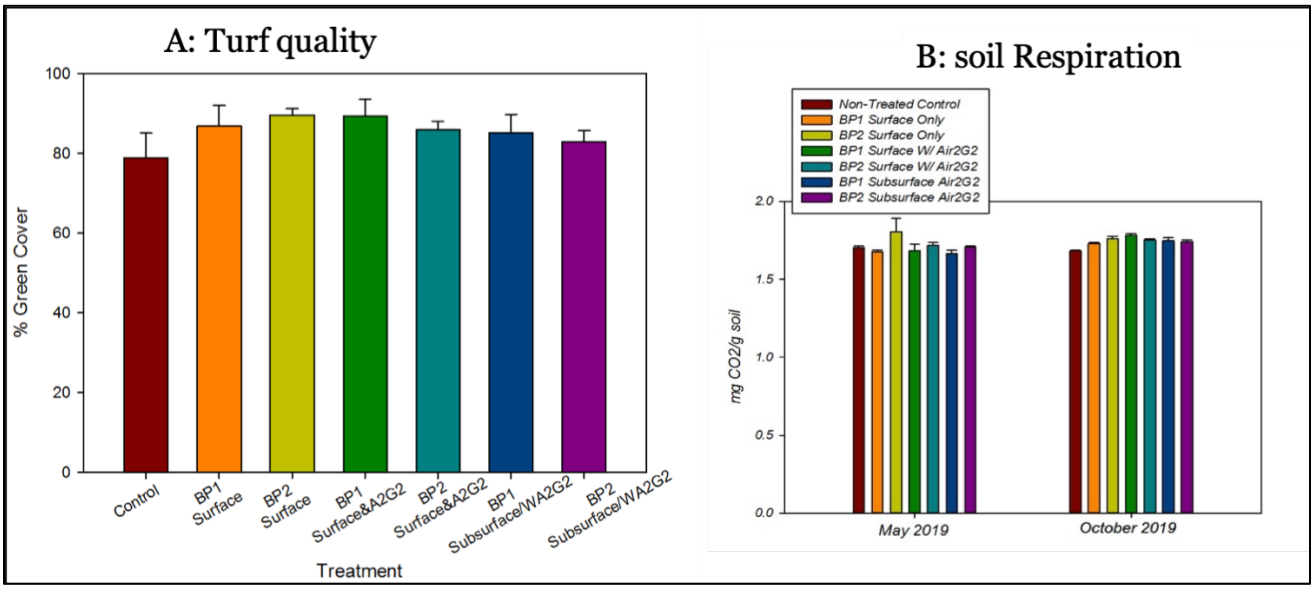

Figure 5: Turf quality (A) and soil respiration (B) in Creeping Bentgrass in Milton, GA.

- Phosphatase and urease: Similar to what we saw in Rivermont, nether phosphatase nor urease activity was significantly impacted by any of the treatments as compared to the Control at the Echelon plots. The only difference was due to sampling time.
- Despite not being significantly different from the Control treatment, subsurface applying BP2 with A2G2 (treatment 7) seemed to result in higher urease activity than BP1 that was surface applied (treatment 4), as shown in May 2019 data (Table 2).

Table 2: Soil phosphatase and urease activities (mean ± 1SE) in Creeping Bentgrass in Milton, GA. Numbers with same letter suffix are not significantly different from each other. Comparison is only valid with in a column.

Treatment	Mean phosphatase activity (µmol P evolved g^{-1} h^{-1})		Mean urease activity (µmol NH_3 evolved g^{-1} h^{-1})	
	May 2019	October 2019	May 2019	October 2019
NTC	0.201ab	0.176abcd	1.27bc	5.48a
BP1 Surface	0.259ab	0.211abcd	0.73bc	5.48a
BP2 Surface	0.309ab	0.129cd	0.82bc	5.23a
BP1 Surface w/ Air2	0.398abc	0.172abcd	0.63c	5.73a
BP2 Surface w/ Air2	0.433ab	0.152bcd	0.73bc	4.98a
BP1 Sub Air2	0.458a	0.074d	1.13bc	4.98a
BP2 Sub Air2	0.343ab	0.072d	1.48b	5.23a

5. Preliminary Conclusion

- Based on the data that have been gathered so far, the biological products have not led to any significant improvement in either turf quality or soil biological health as compared to the Control treatment.

6. Future plans / On-going activities

- Continue treatment applications in both sets of plots (in Milton and Johns Creek) till next year spring.
- Continue taking regular measurements on turf quality and biological soil health indicators at both sites.
- Continue monitoring and rating the plots for disease in response to the inoculations.
- Send Final report by end of summer 2020.

Please let us know if you have any questions.

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Collaborators:

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Mr. Todd Lime, superintendent of Echelon golf club

Appendix

Some pictures from field work at Johns Creek, GA.