The purpose of our study was to characterize perennial ryegrass seedling establishment and growth response using biostimulants alone and with nitrogen.

Over the past two decades, the turf industry has witnessed increased pressure from government agencies and environmental groups over the fate of nutrients in the environment and concern about surface and ground water. Consequently, several new products and technologies have been developed to help turf professionals deal with day-to-day management challenges; one such development is in the introduction of biostimulants. They have particularly generated both enthusiasm and serious skepticism.

Understandingly, in their quest to market their products, biostimulant manufacturers put a lot of emphasis on what their products can do but did not provide evidence to support their claims (see list of common claims in table 1.).

What are biostimulants?

"Biostimulants," often used in plural form, is a broad term that literally means a group of ingredients that stimulate life. This could also be interpreted as a group of compounds that promotes favorable plant responses. Biostimulants have also been described as non-nutritional products.

Others suggested them as materials that stimulate plant growth in minute quantities (Zhang and Schmidt, 1999). The description of biostimulants invariably becomes a discussion about their function and proposed uses in turf management. With these definitions, one could make a good argument that a light, balanced fertilizer application produces biostimulant-like responses i.e., shoot growth, increased nutrient uptake, and photosynthetic translocation.

Obviously, the questions that biostimulants raised are not about definitions but rather lie in their diversified chemical composition. In a recent article, Dr. Karnok compiled about 60 different ingredients listed in 15 biostimulant labels (table 2.)

A lot of the ingredients listed are known organic and mineral substances essential to plants growth and development processes. Common commercial biostimulants contain many, if not, all of the following major ingredients:

- Plant hormones
- Humic substances
- Manure and/or sea kelp extracts

The chemistry of these basic biostimulant ingredients is extremely diverse in nature, origin, synthesis, function, and role in plant-soil ecosystem.

Plant Hormones: Research on plant hormones and their role in regulating plant growth and development processes have been extensively documented. Phytohormones are synthesized in the plant to regulate a multitude of essential cellular and tissue functions including cell enlargement, root initiation, and tissue differentiation.

They are grouped in five major categories: indoleacetic acids (IAA), commonly known as auxins, gibberellic acids, cytokinins, abscisic acids (ABA), and ethylene. Cytokinins are particularly implicated in cell division, morphogenesis (tissue differentiation), nutrient mobilization, and senescence delay.

Auxins promote root and shoot elongation (cell enlargement). Gibberellins promote shoot elongation, regulate seed germination, and seedling establishment. Biosynthesis, metabolism, and action of the different hormones are highly regulated processes. Plants maintain an intricate balance among the various hormones.
Humic Substances: These are complex, organic compounds that can generally be classified into humic acid, fulvic acid, and humin based on their solubility in water as a function of their pH (MacCarthy et al., 1985).

Many earlier studies with field crops showed positive responses to the application of humic substances. Shoot and root growth, seed germination, and seedling establishment are usually reported as direct plant responses to humic substances.

Mechanisms by which humic substances produce such responses remain unclear. However, humic substances, partly because of their complex, organic chemistry, have been demonstrated to improve soil structure, cation exchange capacity (CEC), and microbial activity.

Sea Kelp and Manure Extracts: These products contain a large number of organic and mineral compounds. They are particularly rich in phytohormones, complex organic compounds, vitamins, simple and complex sugars, enzymes, proteins, and amino acids.

Perhaps, sea kelp extracts, also known as seaweed, are best known for their high concentration of cytokinins and auxins.

Common additives to the ingredients described above include nitrogen (N), phosphorous (P), and potassium (K) and iron (Fe). Iron is added as a sulfate or in a chelated form. Potassium is mostly included as potassium sulfate.

In essence, the fact that biostimulants are manufactured and marketed as a complex mixture of ingredients is indicative of the potential functions that they may play in turf management, with plant hormones and humates are the two most important components regardless of their source or extraction process.

Recent research
In a recent study conducted at North Carolina State University, humates were shown to increase root enzymatic activity but produced no effect on visual quality and clipping dry weight. Liu and Cooper (2000) reported a significant root mass increase of creeping bentgrass growing in hydroponic culture using modified Hoagland’s nutrient solution treated with 400 ppm of humic acids. The authors found no similar increases in with 100 and 200-ppm concentration and that incorporating the humates at the depth of four inches produced better root responses than foliar applications.

Hartwigsen and Evans (2000) evaluated the effects of humic acids treatment of geranium and marigold seeds and germination substrates on seedling root development. They found that humic acids treatments significantly increased root fresh weight of geranium and marigold seedlings. Root fresh weight increases were significantly
TABLE 1. COMMON BENEFITS OF BIOSTIMULANTS

<table>
<thead>
<tr>
<th>Reported by manufacturers</th>
<th>Stimulate plants’ immune system</th>
</tr>
</thead>
<tbody>
<tr>
<td>Stimulate plant responses and work in all weather conditions</td>
<td>Produce better color</td>
</tr>
<tr>
<td>Increase profits, cut operating costs, lead to 50% reduction in fertilizer</td>
<td>Result in better performance</td>
</tr>
<tr>
<td>Increase natural plant toxins, repelling pests</td>
<td>Produce deeper roots</td>
</tr>
<tr>
<td>Increase microbial root protection from soil pathogens</td>
<td>Improve stress tolerance</td>
</tr>
<tr>
<td>Increase soil nutrient reserve up to 3000%</td>
<td>Accelerate establishment</td>
</tr>
<tr>
<td>Improve root development</td>
<td>Increases Cation Exchange Capacity</td>
</tr>
<tr>
<td>Build yields</td>
<td>Enhances fertilization and reduces leaching</td>
</tr>
<tr>
<td>Improve taste and shelf-life</td>
<td>Detoxify chemical residues and heavy metals</td>
</tr>
<tr>
<td>Improve drought tolerance</td>
<td>Make urea a long-life nitrogen</td>
</tr>
<tr>
<td>Increases nutrient uptake</td>
<td>Improve seed germination rates</td>
</tr>
<tr>
<td></td>
<td>Increase stomata opening and plant transpiration</td>
</tr>
</tbody>
</table>

higher that than those of nutrient controls, suggesting humic acids have affected seedling fresh weights through mechanisms other nutrient supply.

A study of post-transplant root growth and sapflow of balled and burlapped red maple trees in which three different formulations of humate-based biostimulants were applied, Kelting et al. (1998) found no statistical difference in root mass at harvest as compared to untreated controls and no visual differences in growth or caliper size, although sap flow was significantly increased. Similarly, Harris et al. (1997) found that biostimulants did not significantly benefited summer landscape tree transplants and partly attributed their findings to environmental conditions being favorable for transplant.

Several arguments were also made about biostimulants improving turf tolerance to abiotic stresses especially water stress, which is predictably a major limiting factor in highly managed turf. Water stress impacts several plant metabolic functions, including specifically photosynthesis and photosynthetic transport.

Recently, at Virginia Tech, Zhang and Schmidt (2000) concluded that “hormone-containing substances” and humic acids improved shoot and root growth by increasing concentrations of α-tocopherol in tall fescue and creeping bentgrass grown under low-water regime; an antioxidant implicated in the prevention of water-stress induced damage to the photosynthetic apparatus. In their report, they indicated that exogenous applications of seaweed extracts and humic acids promoted shoot and root growth by influencing antioxidants under low moisture conditions.

These findings were consistent with a similar study on Kentucky bluegrass and with those of Smirnoff (1993) who suggested a close correlation between tissue antioxidant levels and drought tolerance. However, the drought tolerance mechanism of other species remains unclear.

In a study on wheat grown in alkaline soil, humic acids have been shown to reduce phosphorous (P) fixation and increase water soluble P to plants (Wang et al., 1995). This makes a good argument for the potential uses of humate-based biostimulants as aids to nutrient uptake by creating humate-metal-phosphate complex, thus reducing soil fixation of P.

At TruGreen ChemLawn Technical Center, we evaluated two biostimulants supplied by PBI Gordon. The experiment
TABLE 2. INGREDIENTS OF 15 BIOSTIMULANT LABELS

(Karnok, 2000)

- Activated nutrients
- Active Humic acids
- Amides
- Amino acids
- Antioxidants
- Bacteria
- Carbohydrates
- Carbon-rich organics
- Cellulose fiber
- Chelated micronutrients
- Chelates
- Chemical activators
- Complex sugars
- Cultured living microorganisms
- Cyanobacteria
- Cytokinin
- Disaccharides
- Enzymes
- Fermentation materials
- Fungi
- Gibberellic acids
- Growth simulators
- Humic substances
- Humic/Fulvic acids
- Hydrated organic proteins
- Intermediate metabolites
- Invert sugars
- Kelp extract
- Lignin
- Manure extract
- Metabolites
- Micronutrients
- Minerals
- Monosaccharides
- Mycorrhizae
- Natural wetting agents
- N-fixing Bacteria
- Non-ionic wetting agents
- Nutrient broth
- Organic chelates
- Peptides
- PGRs
- Plant extracts
- Plant Hormones
- Plant Nutrients
- Polysaccharides
- Proteins
- Scientifically balanced formulation
  (No ingredients mentioned)
- Sea kelp
- Seaweed
- Secondary nutrients
- Simple sugars
- Soil conditioners
- Sugar acid chelates
- Vitamins
- Wetting agents
- Yeast
- Yucca extract wetting agent

was set up in the greenhouse at our Technical Center outside of Delaware, OH. The purpose of our study was to characterize perennial ryegrass seedling establishment and growth response using biostimulants alone and with nitrogen. Perennial seeds were established in a sand culture. Plants were grown in plastic pots containing air-dried sand. At seeding, pots were treated with biostimulant and/or fertilizer (8-10-10) at the rate of one pound N/1000. Biostimulants were added at the recommended rates using a syringe and then the seeds were covered with a thin layer of sand. The pots
BIOSTIMULANTS

were watered through capillary action. The experimental design was a randomized complete block design with four replicates.

The experiment was carried out for 28 days. Data collected included seedling height, at 7, 10, 15, 21, 28 days after treatment (DAT), color at 21 DAT and 28 DAT, and entire shoot fresh and dry weight, root weight, % root biomass at 28 DAT.

Statistical analyses showed that although biostimulants alone provided better visual seedling establishment, color ratings of pots treated with biostimulant + N, and N alone were significantly higher than those of biostimulant alone or control (see graph). Data on percentage root biomass, shoot, and root dry weights, although visually different, were not statistically significant (picture 1, and 2). This indicates that biostimulants have, at least visually produced better seedling establishment, but did not provide, on their own, better color ratings in the first 28 days after treating.

Are biostimulants needed?

In all of these studies, plants were subjected to controlled nutrient regimes, i.e., all essential nutrients were available to the plants under controlled conditions. The studies do no show the mechanisms involved or what would have been the response, should one or several nutrients were made limited.

For example, Harris et al. (1997) concluded that biostimulants are ostensibly not useful if proper transplant practices are followed with balled and burlapped landscape trees. This is consistent with the principle that under normal growing conditions, plants are self-sufficient and do not respond to exogenous supply of hormones because they are self-sufficient.

University research on biostimulants and their uses in turfgrass management does not fully agree with the claims often made by biostimulant manufacturers especially with regards to reducing fertilizer and pesticide uses. However, there are positive reports that biostimulants show potential uses in turf under stress conditions and promote favorable nutrient mobilization, although the responses to biostimulants may vary depending upon the biostimulant formulation and/or composition and among species.

Future research needs

Biostimulant uses in the turf industry will have to be better defined in order to gain a wide acceptance among turf professionals.

Future research should focus on questions pertaining to their role in seed establishment, turf stand quality, nutrient use and efficiency, abiotic stress tolerance under field conditions.

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