New Benefits of Endophyte-Infected Grasses Emerge

By Parwinder Grewal and Doug Richmond

Endophytes can provide enhanced drought tolerance, summer survival, and insect and disease resistance to grasses.

Endophytes are fungi belonging to the genus *Neotyphodium* that live in the leaves and stems of grasses and are carried from plant to plant only through seed. These fungi do not cause any disease in the grasses, but under most circumstances they are beneficial to the growth and survival of infected plants. This association naturally occurs in perennial ryegrass and several species of fescues, but not in Kentucky bluegrass or bentgrass.

The fungus obtains all food resources from the grass host, but in return provides several benefits to the plant. Research provides evidence of greater persistence and growth of endophyte-containing grasses, relative to plants that lack endophytes.

Endophytes modify how plants acquire and use their available resources and make them more widely adapted to environmental conditions. Endophyte-containing grass cultivars have greater seed survival, germination and establishment than cultivars lacking endophytes. The presence of endophytes confers drought resistance and enhances summer survival of grasses.

Endophyte-containing cultivars also perform better in poor-quality acidic soils and soils with low phosphorus content.

Enhanced insect, nematode resistance

The endophytes provide a built-in systemic pesticide for the plant. Defensive chemicals are produced by the endophyte that are disruptive to numerous animal species capable of using endophyte-free grasses for food.

Endophyte-infected grasses are resistant to sod webworms, chinchbugs, billbugs and numerous other surface-feeding insects. Previous research at The Ohio State University showed that excellent control of bluegrass billbug can be obtained in Kentucky bluegrass lawns overseeded with endophyte-infected perennial ryegrass. This research also established that only about 40 percent of the plants in a turf sward have to contain endophytes to obtain effective control of bluegrass billbug and sod webworm.

Turfgrass insects, nematodes and mite pests that can be managed with the use of endophyte-containing grasses are listed in Table 1 (page 90).

Although endophytes are more effective against above-ground insect pests, below-ground insects and plant-parasitic nematodes are also affected. The fungus and its alkaloids are concentrated in above-ground plant tissues, but as much as 15 percent of the lolines and smaller amounts of ergot alkaloids may occur in tall fescue roots. Ergot alkaloids strongly deterred feeding by the Japanese beetle grubs in laboratory bioassays and reduced their survival and weight gain in stands of endophyte-containing grasses in some field trials. Plant-parasitic nematode populations in the soil have been found to be smaller under endophyte-containing grasses than the endophyte-free grass stands.

Aside from the direct toxic affect that endophyte-infected plants have on many surface feeding insects, endophytes provide the additional benefit of altering insect foraging behavior. Insects such as chinch bugs and sod webworms spend more time moving and less time feeding in stands of turfgrass containing even moderate proportions of endophyte-infected plants.

This increase in movement makes the insects much more vulnerable to predators and pathogens and may be a death knell for newly hatched larvae or nymphs. These neonate insects are equipped with very little in the way of energy reserves, so they must find a suitable food source quickly or perish in the process of searching.

Effects of cultural practices

Levels of endophyte-produced alkaloids depend on several environmental factors. Seasonal variation typically shows maximal levels at the end of the growing season, but peramine in tall fescue shows no seasonal effects.

Temperature, sunlight and rainfall all have an impact on alkaloid production. Our own

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research shows that extreme temperatures repress alkaloid levels relative to the levels between 14 degrees and 21 degrees Celsius in tall fescue and perennial ryegrass.

In a greenhouse trial, we found that the contents of different ergot alkaloids were variable at different temperatures, but there were no clear trends on the effect of temperature on any particular alkaloid between 14 degrees and 25 degrees Celsius, thus supporting the usefulness of planting endophyte-containing grasses in the Midwest. In this trial, we also found that both the survival and weight gain by fall armyworms were significantly lower when feeding on endophyte-containing perennial ryegrass than the nonendophytic perennial ryegrass at all temperatures.

Both nitrogen (N) and phosphorus (P) fertilizers can alter the levels of alkaloids in tall fescue. The application of nitrogen increases the concentration of major ergopeptide alkaloids in tall fescue cultivar Kentucky 31. At the University of Georgia, it was demonstrated that the source of nitrogen is also important, as all concentrations of NO_3 –N increased ergopeptide alkaloid content as opposed to NH_4^+ -N that was effective only at high concentrations.

Ergopeptide concentrations were highest in drought-stressed plants that were fertilized at the moderate or high N rate. Ergot alkaloid accumulation in roots of tall fescue increased linearly with phosphorus availability in the soil, but in the shoots the alkaloid concentration increased with increasing phosphorus availability in the soil from 17 milligrams to 50 milligrams P per kilogram of soil, but declined at 96 milligram P per kilogram of soil.

We found that the levels of ergot alkaloids in endophyte-containing perennial ryegrass and tall fescue are affected by the common cultural practices such as mowing height and mowing frequency. We found that the major ergot alkaloids including ergonovine, ergocristine and ergocriptine significantly increased in tall fescue with increased mowing height from 1 inch to 3 inches (Fig. 1). Increased mowing height reduced masked chafer grub populations at the University of Kentucky.

In another greenhouse experiment, we tested the effect of two mowing frequencies, once a week (weekly) or once every two weeks (biweekly) on ergot alkaloids. The mowing height was 2 inches in this experiment. We found that decreased mowing frequency caused a five-fold increase in the amount of ergovaline and a 2.6-fold increase in ergonovine (Fig. 2).

In both the above experiments we used the fall armyworm as a bioassay insect to detect the impact of our treatments on the resistance of grass plants to insects. We found that as the concentrations of ergonovine in the shoots increased due to less frequent mowing, the dry weight of the fall armyworm decreased.

FIGURE 1

Effect of mowing height on concentrations of major ergot alkaloids in tall fescue shoots after one or two months in the greenhouse at 25 degrees Celsius. Grass was mowed weekly at either 1-, 2- or 3-inch height. Same lowercase letters on the bars indicate no significant differences between mowing height treatments within each month.

Micrograms per gram of tissue dry weight

MOWING HEIGHT	ERGOCRISTINE 1 mo	ERGOCRISTINE 2 mo	ERGONOVINE 1 mo	ERGONOVINE 2 mo	ERGOCRYPTINE 1 mo	ERGOCRYPTINE 2 mo
1 Inch	3.5	4.3	1.5	2.3	1.1	8.5
2 Inch	3.2	3.9	1.6	2.8	2	9
3 Inch	3.8	4.6	1.8	3	3.6	15.3

FIGURE 2

Effect of mowing frequency on concentrations of major ergot alkaloids in tall fescue shoots after two months in the greenhouse at 25 degrees Celsius. Grass was mowed at 2-inch height either weekly or biweekly. Same lowercase letters on the bars indicate no significant differences between mowing height treatments within each month.

Nanograms per gram of tissue dry weight

MOWING FREQUENCY	ERGONOVINE	ERGOVALINE	ERGOCRYPTINE
Weekly	200	3.1	228
Every two weeks	490	14.5	245

Enhanced ability to compete

Endophyte-containing plants are generally more competitive and tend to dominate plant communities over time.

Research in Indiana showed that in mixed perennial ryegrass and white clover swards, the presence of the endophyte in ryegrass resulted in a significant decrease in white clover. Endophyte-containing ryegrass maintained greater cover under severe insect stress, whereas stands lacking the endophyte persisted poorly and were more heavily invaded by weeds.

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Following drought, survival and cover of endophyte-containing tall fescue was significantly enhanced. Reduced cover of stands lacking the endophyte allowed other grasses and dicots to increase. These results indicate that the abundance and diversity of other plant species may be reduced by competition with endophyte-containing grasses.

Our research demonstrates the competitive ability of endophyte-containing grasses could substantially contribute to reduction in weed invasion and establishment in lawns.

Allelopathic effects of tall fescue on red clover and birdsfoot trefoil were first demonstrated in the 1970s. Our own recent research shows that endophyte-containing tall fescue and perennial ryegrass are often better able to compete against common weed species. In a field study on newly established plots, we observed that the population of crabgrass was lower in endophyte-infected stands of perennial ryegrass compared to uninfected stands.

Likewise, the density of plantains (buckhorn and common) was consistently lower in stands of endophyte infected tall fescue compared with uninfected stands. These plots were under minimal management, received no fertilizer or pesticide inputs, and were mowed at 3.5-inch heights.

TABLE 1

Major insect, mite and nematode pests of cool-season turf that can be potentially managed with endophyte-infected grasses.

INSECTS

Annual bluegrass weevils, *Hyperodes spp*
Armyworm, *Pseudaletia unipuncta*
Bluegrass billbug, *Sphenophorus parvulus*
Chinch bug, *Blissus leucopterus leucopterus*
Hairy chinch bug, *Blissus leucopterus hirtus*
Black cutworm, *Agrotis ipsilon**
Bronzed cutworm, *Nephelodes minians*
Fall armyworm, *Spodoptera frugiperda**
Greenbug, *Schizaphis graminum (Rondani)*
Bluegrass webworm, *Parapediasia teterrellus*
Cranberry girdler, *Chrysoteuchia topiaria*
Larger sod webworm, *Pediasia trisectus*
Vagabond crambus, *Agriphila vulgivagellus*
Japanese beetle, *Popillia japonica**
Southern masked chafer, *Cyclocephala lurida**

MITES

Clover mite, *Bryobia praetiosa*
Winter grain mite, *Penthaleus major*

PLANT PARASITIC NEMATODES

Ring nematode *Crionemoides spp.*
Spiral nematode *Helicotylenchus spp.*
Lance nematode *Hoplolaimus spp.*
Lesion nematode *Pratylenchus spp.*
Stubby root nematode *Tylenchorhynchus spp.*
Pin nematode *Longidorus sp.*
Dagger nematode *Xiphinema sp.*

*Represents species that can feed and develop on endophytic grasses, but have slower development than those feeding on nonendophytic grasses

Weed management

Aside from providing protection from insect pests, endophytes can provide a measure of protection against other important turfgrass problems that are not so obviously related.

Although it's a fact that insect feeding can reduce the vigor and persistence of turfgrass plants, we rarely make the connection between this kind of damage and other ensuing problems such as weed invasions. In fact, we tend to think of weeds and insects as separate and independent concerns in turfgrass management and, to a large extent, our approach toward research and extension reflects this way of thinking.

When turfgrasses are attacked by insects, their ability to compete with the ever-present pressure from encroaching weeds is degraded. Even worse, when insects cause mortality of turfgrass plants, the new occupant of the formerly turfgrass covered site will likely be an undesirable plant.

In a simple study, we observed weed invasion into stands of Kentucky bluegrass suffering from different levels of damage caused by the bluegrass billbug. At the undamaged site, few weeds were ever present, and none of these weeds were able to become established. However, at the site where moderate billbug damage was apparent, weeds, such as dandelion, black medic and crabgrass, were a significant problem.

But what if the above-mentioned sites had been planted with endophyte-enhanced turfgrasses? For one thing, insects that feed on the above-ground portions of turfgrass plants (billbugs, armyworms, cutworms, sod webworms and chinch bugs) are much less likely to cause significant damage to endophyte-enhanced grasses.

In a series of greenhouse studies, we observed competition between dandelion and two species of cool-season turfgrass (tall fescue and perennial ryegrass) infected or uninfected by fungal endophytes. When fall armyworms were present, endophyte-infected grasses were much stronger competitors against dandelions. Under these circumstances, endophyte-infected turfgrasses produced more tillers and had greater biomass than uninfected plants.

Conversely, dandelion plants had fewer leaves and lower biomass when competing against endophyte-infected plants. These studies indicate that because endophyte-enhanced turfgrasses are better able to defend themselves against foliage-feeding insects, they are also much more vigorous competitors against some of our most common weed species. Endophyte-enhanced grasses not only provide the direct benefit of lowering populations of insect pests, but they also provide an indirect benefit by resisting weed invasion.

System-wide effects of endophytes

It's obvious that fungal endophytes can provide a range of benefits for turf. However, it is important to recognize that the benefits of insect resistance, competitiveness against weeds and broader range of adaptation are not completely independent of one another.

For instance, we have conducted controlled studies that demonstrate how resistance to insects also provides a competitive advantage against weeds. Since endophyte-infected grasses are not preferred by insect, they suffer much less damage and, as a result, are much stronger competitors against dandelions and other common weed species.

Likewise, since endophyte-infected plants are more efficient at acquiring nutrients from the soil, they are less likely to become overrun by weeds in times of drought or when nutrients are in short supply. This phenomenon of multiple, simultaneous and interacting benefits provides the basis on which a systems-oriented approach to turfgrass management is founded.

Realizing the benefits of endophyte-infected grasses does not always require complete renovation. Since only moderate proportions of endophyte-infected grasses are necessary to provide resistance against many insects, overseeding preexisting stands is a simple and effective option.

Previous research at The Ohio State University indicates that overseeding stands of Kentucky bluegrass with endophyte-infected perennial ryegrass at a rate of 1 pound or 2 pounds per 1,000 square feet (using a slicer-seeder) can significantly alter the composition of the turfgrass stand within a short period. Overseeding in the fall will provide resistance to billbugs, chinch bugs and sod webworms the next year. Over time, perennial ryegrass will outcompete the Kentucky bluegrass and will gradually take over the stand. As a result, both insects and weeds will be much less of a problem.

Seed storage

As endophyte is present in the seed, it is very important to store the seed in a cool and dry place. Temperatures exceeding 37 degrees Celsius for one to two weeks can eliminate the endophyte from the seed while the seed may still be viable. Therefore, the seed should not be stored in the garage because it may get too hot in the summer.

It's also important to use fresh seed that has been stored under cool conditions. A grow-out test can also be performed to confirm endophyte viability in the seed.

Use of endophyte-containing grasses offers a real, sustainable solution to turfgrass pest management. Endophyte-containing grass will have mitigating effects on pest problems including weeds, insects, diseases and plant-parasitic nematodes. Thus, it provides a turf system that is more resistant to pest invasion and establishment.

Our ability to boost alkaloid (toxin) levels in endophyte-containing plants by simple cultural practices such as reduced mowing frequency, increased mowing height, increased nitrate nitrogen and phosphorus (coupled with reduced irrigation frequency) opens doors to obtain enhanced benefits from planting the endophyte-containing grass cultivars. The resulting lesser use of chemical pesticides and surfactants would also favor colonization and establishment of natural enemies, parasites, predators and pathogens of pests.

The use of endophyte-containing grasses in turf systems is practical as endophyte-containing grasses can be easily incorporated into pest management systems by overseeding in the existing courses or by establishing new courses containing mixtures of endophyte-enhanced and endophyte-free grasses.

Grewal is an associate professor of entomology at the Ohio State University. Richmond is a post-doctoral research associate in Grewal's laboratory.