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The Good & Bad of Endophytes

Significance of Fungal Endophytes in Turf and Forage Grasses

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Introduction

The field of endophyte research is relatively new even though the endophytic fungi have been known to occur within certain grasses for over one hundred years. These endophytic fungi grow systemically within the plant tissues and produce no external signs of infection. In 1977, Bacon et al. made the first association between an endophyte in tall fescue and certain maladies of cattle comprising what is known as "summer syndrome." Since then, research on the grass endophytes has been greatly accelerated, both in the beneficial and detrimental aspects of fungal endophytes in turf and forage grasses. These studies include investigations into the symptomatology of several distinct animal toxicoses, biological control of turfgrass insects, and research on the associated toxins. Information about fungal endophytes, their host range, distribution, biology and toxicology, is currently of importance to agronomists, plant pathologists, professional turf managers, seed producers and processors, animal scientists, entomologists, I.P.M. specialists and soil conservationists.

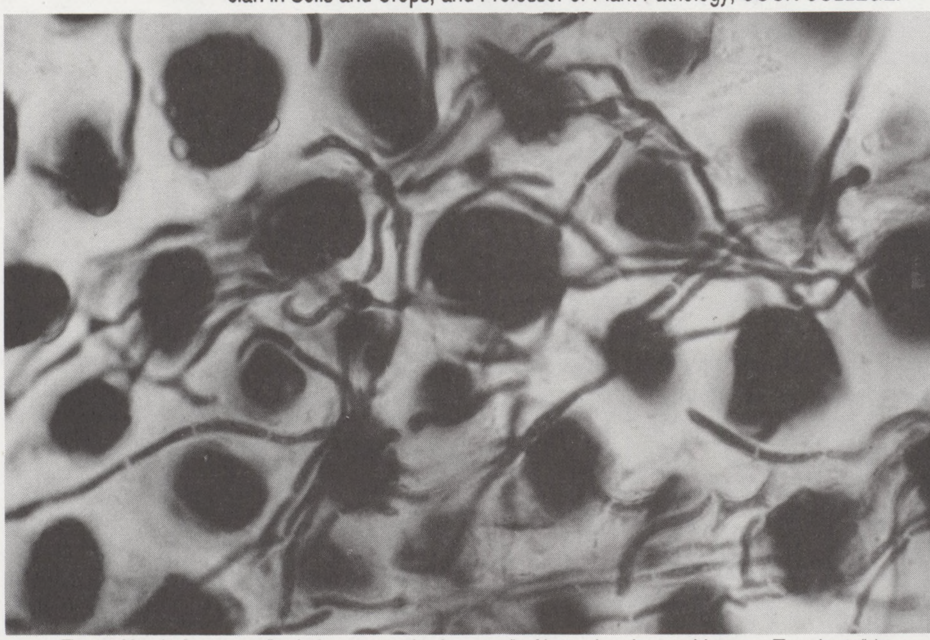

Figure 1. Here the fungus mycelium is shown among the aleurone cells of the seed coat in perennial ryegrass. These intercellula threads grow systemically in the grass plant in the leaf sheath, pith of the culm, and the seed coat.

Endophytic Fungi

Five species of Acremonium are known to infect turf and forage grasses. These are A. typhinum, A. loliae, A. coenophialum, and two recently reported species, A.. huerfanum and A. chisosum. The genus Acremonium is classified in the Fungi Imperfecti in the family Moniliaceae. With the exception of A. typhinum, no sexual stages of these fungi have been produced in artificial culture or found in nature. No external symptoms or signs of these fungi have been reported, therefore the grasses infected with them are "symptomless." To date, no measurable detrimental effect of the fungi on their grass hosts have been observed.

Endophytic fungi are transmitted through the maternal plant in seed. Transmission through the pollen is not known to occur. The fungal mycelium passes into the seed by going up the stem and then into the flower. In mature seed, the mycelium is found chiefly between the scutellum and aleurone layer. Infection of the young plant begins during germination and is very complete. However, the fungus is present only in the aerial parts of the plant. Higher concentrations are found in the leaf sheaths, pith and seed, and lower concentrations in the leaf blades.

The endophyte is also disseminated when grass plants are clonally propagated. Techniques for the artificial inoculation of endophyte-free grasses with these fungi have been developed but have very limited usage (Siegel et al., 1985).

Endophyte Hosts

Endophytic fungi are known to infect a wide range of grass hosts including both forage and turf grasses. White (1987) examined over 800 herbarium specimens in 93 genera of grasses. Using microscopic examination of stained pith tissues to detect endophytic mycelium, he found the fungi to be present in 43 grass species in 11 genera. These genera include Agrostis, Bromus, Cinna, Digitaria, Elymus, Festuca, Lolium, Melica, Poa, Sitanion and Stipa. Note that Cynodon (bermudagrass) and Zoysia (Japanese lawn grass) are not currently numbered among the hosts of Acremonium endo-

continued on page 4

OPINIONS AND COMMENTS

Annual Bluegrass Herbicides

Several herbicides are being sold for annual bluegrass control. Each is quite different chemically. Unfortunately, there is too little research on these by the State Agricultural Experiment Stations. Refereed research publications are very limited. I do not doubt these chemicals possess herbicidal action on annual bluegrass, but a big and complicated step is required to determine how delicate bentgrass turf will react to a chemical in a variety of locations over a period of years. Learn all you can from others and read any authentic research reports that become available on this group of herbicides. Some products have little formal testing, whether they are older or well known. Surely they differ in success. Hopefully, they all might make a long-term contribution, but the odds are against this. If you try one or more of these herbicides, do so with a cautious and circumspect approach. Several years of trial may be necessary to evaluate the long-term results.

❒ REE

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A Science of Trade-offs?

After the last issue of **Greenworld**, I read several references about mixing politics with science. I hope political and social pressure on scientific direction and conclusions continues to diminish as our world is developing greater interdependence with science. Maintaining scientific integrity need not be in conflict with other things — just a guide to reality. In any mixing with politics and social desires, science provides boundaries on what can be done. For example, height of cut on turf is often below optimum for the grass and climate, but we decide to pay and suffer for this violation in satisfying our turf use or personal preferences. We must remain aware of the truth that respecting the optimum cut makes turf growing easier. Without it, we lose perspective on maintenance standards. ❒ REE

—ABSTRACT—

Turf Cultivation Effects on Preemergence Crabrass Control

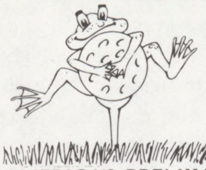
B.E. Branham & P.E. Rieke,1986
Agronomy Journal 78: 1089-1091

Cultivation is an important cultural practice used to relieve soil compaction and improve turf rooting. Cultivation is often avoided during periods when weed seeds germinate for fear of disrupting the preemergence herbicide and subsequently decreasing annual grass control. The purpose of the study was to compare benefin, bensulide, and DCPA control of crabgrass with untreated check plots. Additionally, cultivation treatments consisting of core cultivation, one pass or three passes, or vertical mowing, and a control were applied at the time of herbicide application of 4 weeks after herbicide application. Crabgrass populations were evaluated throughout the summer to determine the effects of the herbicide and cultivation treatments. Neither cultivation nor time of cultivation had a significant effect on the degree of crabgrass control afforded by the herbicides. The only significant difference in the study occurred when comparing the herbicide-treated plots with an untreated control. Herbicide-treated plots had less than 10% crabgrass cover compared with 30% or more crabgrass cover in turf areas that received no herbicide. Data indicated that cultivation operations can be safely performed in the spring without disrupting the preemergence herbicide barrier.

The authors stress that the study was conducted on turf maintained as a fairway with a very high turf density. Cores were also allowed to air dry and were dispersed over the turf. Thus, a herbicide barrier was maintained. The authors conclude that as long as the correct herbicides are applied properly, cultivation should not significantly affect herbicide performance.

[Editorial Comment — *A few turf cultivation treatments before and after preemerge treatments of approximately ten years ago in New Jersey showed similar results. Considering all the possible conditions of seed abundance, seed location in the soil profile, and cultivation techniques, it is difficult to conclude positively that cultivation never adds to crabgrass problems. Cultivation during periods of rapid turfgrass growth and far in advance of the crabgrass germination period will enhance healing of the turf prior to crabgrass germination. Maintaining a dense turf is the best defense against weed pests. Likewise, proper selection and application of herbicides is necessary for satisfactory control when chemical weeding is required. The study above demonstrates that as long as preemergence herbicides are active, turf cultivation may not contribute to increased crabgrass problems. However, we should remember that weed problems may increase when cultivation and herbicide treatments are improperly timed or applied.*]

❐ **RHW** and **REE**



continued from page 1

The Good & Bad of Endophytes

phytes. Perhaps the two most important grass hosts of these fungi are tall fescue (Festuca arundinacea) and perennial ryegrass (Lolium perenne). These are important turfgrasses and major pasture grasses for animals such as cattle and sheep. They are also the grasses implicated in animal toxicoses.

Geographic Distribution

The endophytes are known to be distributed throughout many areas of the world. White (1987) has reported that the endophytes were present in grasses collected from Africa, Argentina, Canada, Europe, India, Mexico, New Zealand and the United States. Among the grasses examined from the continental United States, many were "native" species. The presence of Acremonium endophytes in many of our native grasses suggests to us that not all of the known grass endophytes were introduced to North America from Europe. In many of the grasses, the endophytes, like their hosts, may well be indigenous. In fine fescues (Festuca spp.), however, Saha et al. (1987) presented evidence that the endophytes in those grasses were of European origin.

Detection

The fungal endophytes can be detected in an infected plant using several techniques. The most common and most direct method is the staining of plant tissue and subsequent examination under a light microscope. The tissues examined include leaf sheaths and stem pith scrapings. Seeds can also be stained and examined microscopically for the presence of Acremonium mycelium. An alkali-soak-stain method was developed at Rutgers that is very useful for preparing grass seeds for microscopic examination (Halisky & Funk, 1984).

The fungus can also be detected using an enzyme-linked immunosorbent assay (ELISA) which can detect the fungus in very low quantities (Siegel et al., 1987). However, ELISA is fairly expensive, requires specialized equipment, and can only give a relative level (high or low) of infection, rather than a precise level of endophyte present.

Some endophytes are associated with the production of certain compounds which can be detected through chemical analysis. For example, tall fescue can be tested for the presence of pyrrolizidine alkaloids such as N-formyl loline and N-acetyl loline which are found in infected tall fescue grasses. Similarly, perennial ryegrass can be tested for the presence of lolitrem toxins (Siegel et al., 1985).

Animal Toxicoses

Endophytic fungi have been implicated as the cause of several maladies of livestock grazing on infected grass. Cattle grazing on infected tall fescue are often affected by what is commonly known as "summer syndrome." This is characterized by a general decrease in animal performance. This syndrome is sporadic and is more frequent during periods of high temperature (Hemken, 1983).

Another common malady is "fescue foot," which can result in the loss of the feet, ears and tail switch due to gangrene, and is similar to ergot toxicity. "Agalactia," which is more common in horses grazing on infected tall fescue, involves a thickening of the placenta and the production of weak foals, along with a decrease in milk production (Hemken, 1983).

Endophyte toxicity is also the explanation given for the condition known as "ryegrass staggers." This malady most commonly affects sheep in New Zealand, but also cattle, horses and deer grazing on infected perennial ryegrass. The symptoms include tremors and uncoordinated postural reflexes and movements. Generally, ryegrass stagers is worse during periods of severe heat and drought stress, especially when overgrazing occurs. Sheep losses can be considerable (Mortimer et al., 1984).

Insect Control

The endophytic fungi have been shown to aid grasses in tolerating various environmental stresses, particularly insect damage. In 1982, Prestidge and co-workers in New Zealand found that populations of Argentine stem weevil were much lower in plots containing endophyte-infected ryegrasses than in those containing endophyte-free ryegrasses. There was less injury occurring in endophyte-infected plots due to selective feeding by the Argentine stem weevil. Furthermore, they found that endophyte-infected grasses became more dominant in the plots as selection pressures favored them over uninfected grasses (Prestidge et al., 1982).

Working with turf research plots in New Jersey, Funk and co-workers (1983) demonstrated that endophytic fungi were associated with perennial ryegrass resistance to sod webworms. In this study, resistant and susceptible cultivars of perennial ryegrass were analyzed for the presence of endophytic mycelium using both ELISA and microscopic examination. They found that the grasses showing resistance to the sod webworm contained high levels of endophyte, while susceptible grasses contained no endophyte.

Recently, Siegel et al. (1987) compiled a detailed literature review on grass endophytes. They presented a table showing 12 insect species known to be suppressed by endophytes in hosts such as tall fescue (TF), chewing fescue (CF) and perennial ryegrass (PRG). These insects are: sod webworm (PRG), Argentine stem weevil (PRG,TF), billbug (PRG), fall armyworm (PRG,TF), house cricket (PRG,TF), oat-bird cherry aphid (TF), greenbug aphid (PRG,TF), milkweed bug (TF), chinch bug (CF), black beetle (PRG), sharpshooter leafhopper (TF), and corn flea beetle (TF).

Toxins

The endophytes have been found to produce certain chemical compounds or toxins which are thought to be responsible for both the animal toxicoses and the insect resistance. Tall fescue, infected with the endophyte, Acremonium coe-

continued from page 4

The Good & Bad of Endophytes

nophialum, has been found to contain certain pyrrolizidine alkaloids, particularly N-formyl loline and N-acetyl loline. Ergot alkaloids were also found. Endophyte-infected perennial ryegrass produces neurotoxic tremorgens called lolitrems and an alkaloid called peramine. The lokitrems are implicated in ryegrass staggers. These compounds along with peramine are also thought to be the factors responsible for the enhanced insect resistance (Bush et al., 1982; Siegel et al., 1987).

It has also been found that endophytes are responsible for other kinds of toxicoses. Melica decumbens, which is found only in South Africa, is known as "dronk grass." This grass is so named because of the severe staggers condition which affects cattle grazing on it. This grass was found to contain an endophyte. Similarly, Stipa robusta, which occurs in parts of North America, is called "sleepy grass" because of the narcotic effect it has on grazing animals. It too contains an endophyte. It seems possible that an endophyte may also be responsible for the unpalatibility of grasses such as Digitaria insularis or "sour grass." Further studies are needed to confirm the role of fungal endophytes in these cases (white, 1987).

Other Diseases

Some endophytic fungi, in particular Acremonium loliae and Acremonium coenophialum, have recently been shown to suppress growth and sporulation of other fungi in laboratory cultures (White 1985; Bayaa et al., 1987). Among the fungi suppressed were turfgrass pathogens such as Drechslera, Sclerotinia, Rhizoctonia, Sclerotium, and Curvularia. In addition to these turfgrass-infecting fungi, the growth of five other plant pathogenic fungi was inhibited as well as that of three saprophytic fungi. This antibiotic phenomenon appears to be toxin related and may well be mediated by the same toxins associated with the animal toxicoses and insect resistance. However, further research is needed to substantiate such relationships.

Our preliminary observations of turf plots containing grasses with and without endophytes do not support an endophyte-disease resistance relationship at this time. In field trials, diseases such as rusts, dollar spot, and brown patch appear to be equally prevalent on both endophyte-containing and endophyte-free cultivars and selections under turf maintenance. Therefore, the inhibition of pathogenic fungi in laboratory cultures by Acremonium endophytes does not appear to extend to a parallel phenomenon in the field.

Additional Benefits

It seems quite apparent that endophytic fungi impart a number of other beneficial effects on the grass hosts that contain them. Research has consistently shown that endophyte-infected grasses persist better than do non-infected grasses. Funk et al. (1985) found that in many stands, endophyte-infected grasses become more dominant as selection pressure favor them over non-infected grasses. One reason that endophyte-containing grasses are able to withstand more environmental stress may be due in part to increased tillering as has been demonstrated in bentgrass plants by Bradshaw (1959). Infected plants may be better able to withstand heat and drought, and recover more quickly from injury. The increased density occasionally found in endophyte-infected plants may also help to reduce broadleaf weed and crabgrass invasion since there is less space for the weeds to become established. Thus, endophyte infected grasses may be more vigorous than their uninfected counterparts and tend to persist longer.

Perhaps the most important beneficial factor, however, is that the endophytic fungi, with particular regard to the insect resistance they impart, provide a very attractive means of biological control. They fit in well with I.P.M. programs and the resistance is long-term, especially since the endophyte is transmissible to succeeding generations. Growing endophyte-infected plants which will produce infected seed is relatively easy, and infected seeds are convenient to use by the consumer interested in turfgrasses. Finally, turfgrass endophytes may serve to reduce the amount of chemical pesticides introduced into the environment, and this makes endophyte-containing grasses very attractive.

Grasses with Endophyte

It is important to realize that the level of endophyte in grasses is not constant. The level will vary between grass species, cultivars and even between various seed lots. For example, we microscopically examined over 700 seed sources for the level of endophyte. Approximately 60.2% of the perennial ryegrass, 58.8% of the tall fescue and 20.3% of the fine fescue seed sources were infected. [See Table 1] Kentucky bluegrass, in contrast, contained no endophyte, although other species of Poa are known to be infected.

Endophyte content becomes important when considering grass usage. A selection or cultivar with a high endophyte content would be well suited to a lawn environment, whereas it may cause animal toxicoses if used in a pasture. Therefore, it is important to know which cultivars and selections contain endophyte and to what degree they contain them. In this way, the fungus can be managed wisely

continued on page 6

continued from page 5

The Good & Bad of Endophytes

and efficiently.

It is also important to know that the endophyte will lose its viability after storage for one or two years at ambient temperatures. Cool, dry storage conditions, on the other hand, will prolong viability. Consequently, it is important to determine how old the seed is and how it was stored prior to use (Siegel et al., 1985).

The following table shows varieties and selections of ryegrass (Lolium) and fescues (Festuca spp.) with 70 to 100 percent of the seeds in some seed lots infected with endophyte mycelium. These levels of infection were determined using the alkali-soak-stain method of seed processing followed by microscopic examination of the stained seeds.

Table 1
Cultivars and Selections of Turfgrasses with 70 - 100 % of Seeds in Lots Carrying Endophytic Mycelium.

Perennial Ryegrasses
All *Star, Citation II, Commander, Cowboy, Dandy, Dasher II, Omega II, Pennant, Pick 715, Pinnacle, PST M2E, PST 2PM, Regal, Repell, Sherwood, SR-4000, SR-4100.

Tall Fescue
Chesapeake, Clemfine, Kentucky 31, GA-82, DBC, PE 7E.

Hard Fescue
SR-3000

Chewings Fescue
Beauty, Longfellow

Sheeps Fescue
R315, R571, R572.

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