## Endophyte Toxins in Grass Seed Fields and Straw Effects on Livestock

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Grazing animals on grass seed fields and feeding grass seed straw can be advantageous to both livestock producers and grass seed growers. Problems may develop, however, if livestock consume varieties of tall fescue and perennial ryegrass that are intended for use as turf. Some turf varieties are infected with an endophyte, which produces toxins harmful to livestock.

The phaseout of grass seed field burning in Oregon's Willamette Valley during the 1990s resulted in more grass seed straw being baled for use as livestock feed, both locally and for export to Japan, Korea, and Taiwan. In several instances in Oregon, livestock producers have unknowingly created serious herd health problems by attempting to use high-endophyte straw as a direct replacement for hay.

### What is endophyte?

Endophyte is a fungus that lives within a plant (*endo* = inside + *phyte* = plant). The relationship between a grass and its endophyte is symbiotic; that is, they both benefit. The grass provides nutrients, and the endophyte produces toxins that fend off insects, diseases, and grazing animals. Endophyte also helps the grass tolerate drought and other environmental stresses. Since endophyte does not affect the appearance of the grass plant, its presence can be detected only by laboratory analysis.

Some grass varieties grown for turf seed have high levels of endophyte infection. The reason is that turf breeders select for the pest resistance and other good qualities that endophyte-infected plants have, without worrying about the effect of the



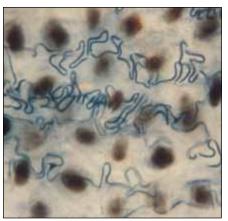


Figure 1.—Endophyte fungus hyphae between the seed coat and the aleurone layer of a grass seed. The dark spots are protein granules in the aleurone cells. (Photo courtesy of Ron Welty (retired) USDA-ARS National Forage Seed Production Research Center)

grass on grazing animals. Breeders of forage varieties, on the other hand, have been deliberately selecting out infected plants since the late 1970s, when the connection between endophyte in tall fescue and a livestock disorder called *fescue toxicosis* was discovered.

The forage varieties of tall fescue and perennial ryegrass seed produced currently in Oregon are endophyte-free or have very low endophyte levels. Turf varieties, however, are an increasingly larger segment of the total grass seed production. Also, some new turf varieties are higher in endophyte than older ones. Therefore, the risk to livestock from Oregon grass seed fields and straw has increased.

Endophyte is transmitted only through the seed, and its entire life cycle takes place inside the plant tissues (Figure 1). A plant does not become infected from its neighbors. Therefore, a stand of a

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noninfected variety will remain noninfected. If a field is overseeded with an infected variety, only the new plants will be infected. A stand of an infected variety cannot be cured with an application of fungicide.

Different species of endophyte fungus infect tall fescue and perennial ryegrass. The fungus infecting tall fescue is *Neotyphodium coenophialum*. It produces 32 (identified so far) toxic alkaloids, including 17 ergoalkaloids that affect livestock. The principal toxin is ergovaline. The other alkaloids affect insects primarily and mammals only to a minor extent. The fungus that infects perennial ryegrass, *Neotyphodium lolii*, produces 16 alkaloids. Six lolitrem alkaloids affect livestock, while the others affect primarily insects. The principal livestock toxin is lolitrem B.

#### **Disease symptoms in livestock**

Fescue toxicosis in livestock is caused by ergovaline and other ergopeptides. These toxins are vasoconstrictors. They constrict the blood vessels and reduce circulation to the body extremities. This interferes with animals' ability to regulate body temperature, causing conditions called *fescue foot* in cold weather and *summer slump* in hot weather.

Fescue foot is characterized by dry gangrene (tissue death) in the extremities. Animals with this condition show lameness and swelling in the legs, followed 2 or more weeks later by loss of the tips of tail or ears and sloughing of the hooves. A 10- to 20-day period of feeding on endophyte-infected tall fescue is required before clinical signs appear.

Summer slump is characterized by hyperthermia (elevated body temperature). Animals spend less time grazing and more time standing in water or shade in order to cool off. Other clinical signs of fescue toxicosis include reduced food intake and poor weight gain, lower pregnancy rates, and decreased milk production. Poor livestock performance is more pronounced when temperatures exceed 87°F. The reduction in weight gain and hyperthermia in cattle may last up to 6 weeks after they are removed from endophyte-infected pasture.

Horses are especially prone to developing serious reproductive abnormalities from ergovaline, including failure to come into heat, early-term abortions, prolonged pregnancies, difficult births, retained placentas, poor udder development with little or no milk production, and poor foal survival. Pregnant mares that are removed from endophyte-infected pasture 1 month before foaling usually recover from fescue toxicosis and have normal foals. However, milk production may be decreased.

In the Northwest, there have been sporadic occurrences of fescue foot in cattle and sheep herds during the winter. In the Southeast, where summer temperatures are higher, summer slump is common. Reproductive problems in horses occur in both the Northwest and the Southeast. With more than 35 million acres of mostly endophyte-infected tall fescue pasture in the southeastern states, fescue toxicosis caused by endophyte is the number one large animal toxicity problem in the United States. Livestock losses are estimated by the U.S. Department of Agriculture at nearly \$1 billion per year.

*Ryegrass staggers* is caused by lolitrem B and other lolitrem alkaloids. These toxins are tremorgens. They cause muscle weakness, tremors, and spasms. Most affected animals show no clinical signs unless they are excited. When they try to run, however, they experience problems ranging from trembling to severe incoordination and falling down. In Oregon, this condition is most common in sheep grazing endophyte-infected perennial ryegrass as their only feed. A 7- to 14-day exposure is required. The signs usually disappear 2 to 3 days after animals are removed from the feed, but sometimes can last as long as 2 weeks.

### Is there a safe level?

Experiments and case studies conducted at the Oregon State University College of Veterinary Medicine and elsewhere have determined threshold levels of ergovaline and lolitrem B in the diet (Table 1). Clinical disease is not seen at toxin levels below the threshold.

It is important to emphasize that these thresholds refer to the level of toxin in the total diet, not in single feed components. Forages with higher

# **Table 1.**—Threshold levels of ergovaline and lolitrem B in the diet that produce clinical disease.

	Ergovaline parts per billion	Lolitrem B parts per billion
	(ppb)	(ppb)
Horses*	300-500	not determined
Cattle	400-750	1,800-2,000
Sheep	500-800	1,800-2,000

\*Except for mares in the last 60 to 90 days of pregnancy, when the threshold is zero.

toxin levels may be fed safely, as long as they are diluted with other feedstuffs.

Threshold levels vary because environment and stress also play a role in the development of clinical disease. Most of the grass seed straw fed to livestock in Oregon is fed in the winter, and extreme cold in some localities increases the severity of the problem. Frostbite can be a complication in fescue foot. If animals are also in poor nutritional condition, the problems are even more severe. This can result from feeding straw without appropriate supplementation, since straw is usually less nutritious than hay.

It once was thought that ergovaline was present only in tall fescue. However, a survey of Willamette Valley grass seed straw in 1992 found that endophyte-infected perennial ryegrass contained ergovaline as well as lolitrem B. Ergovaline is now known to be one of the alkaloids produced by *Neotyphodium lolii*.

Another OSU study in 2001 found that, on average, the ratio of lolitrem B to ergovaline in perennial ryegrass straw is 10 to 1. A perennial ryegrass with 2,000 ppb lolitrem B, for example, would have only 200 ppb ergovaline. In this grass, the ergovaline would not be a problem, except for pregnant mares in the last trimester. For this reason, animals fed endophyte-infected ryegrass almost always show ryegrass staggers before symptoms of fescue toxicosis.

### Avoiding endophyte toxins

Pastures planted with certified seed of Oregongrown forage varieties of tall fescue or perennial ryegrass are safe to graze or cut for hay. For planting a new pasture, using seed that carries the Oregon Department of Agriculture "Endophyte Fungus Tested" green tag will provide additional assurance. This tag verifies that the seed meets the Oregon forage grass standard of 5 percent endophyte or less. To keep pastures endophyte free, they should not be overseeded with seed of turfgrass varieties.

The endophtye fungus hyphae are concentrated at the growing points in a grass plant (the meristematic tissue). The fungus first is found at the base of the plant in the leaf sheaths. As the grass enters its reproductive phase, the endophtye moves up from the base as the stem elongates, into the seedheads, in order to be transmitted to the next generation. The highest concentration of ergovaline is found in the seedheads. Therefore, feeding seed screenings from unknown grass seed fields is risky. Ergovaline toxicity is not reduced by pelleting the screenings.

With both tall fescue and perennial ryegrass, grazing the grass short carries more risk of toxicity, because of the endophyte concentration in the basal leaf sheaths. The crowns of tall fescue have around twice the amount of ergovaline as the tops. Thus, grazing the crowns in a grass seed field following seed harvest is riskier than grazing the field later in the winter.

In both tall fescue and perennial ryegrass, the endophyte fungus loses viability in seed that is stored 18 months or more, especially if it is stored at high temperature and high humidity levels. The percent of infection in old seed and in plants grown from old seed is reduced.

Ergovaline, however, is still present after several years of storage. Consequently, storing infected grass seed straw or screenings doesn't make them safer to feed. Making silage does not reduce ergovaline toxicity, but ammoniating straw reduces the ergovaline level by approximately 50 percent.

Ergovaline levels may vary among different fields of the same grass variety with the same level of endophyte infection. Also, the ergovaline level in the same field varies at different times of year and in different years. Increased nitrogen fertilization can increase the ergovaline level. More ergovaline is produced when plants are growing under stress. Research at OSU in 1991–1992 showed that the ergovaline content in straw from the same plants was 55 percent less in 1992 than in 1991, due to the stress from higher than normal spring rainfall the first year.

An OSU study in 2001 measured the ergovaline in a high-endophyte turf-type tall fescue field (77 percent infection) in different months and in different parts of the plant. Quantities of ergovaline were highest during September and October, and lowest in December and January. The September-October averages were approximately 2,000 ppb in the seed, 900 ppb in the crowns, and 460 ppb in the tops and stems. Since the sheep in this field were rotated frequently and grazed mostly the tops of the grasses, their diet was estimated to contain approximately 460 ppb ergovaline. None of these animals developed clinical signs of fescue toxicosis. Ergovaline levels in the winter months were approximately 500 ppb in the crowns, 200 ppb in the stems, and 75 ppb in the tops.

Since the toxin level within endophyte-infected forage is highly variable, testing for the toxins is the best way to decide how, or if, a feedstuff should be used in a livestock diet. Testing is strongly recommended for grass seed straw of unknown variety, particularly if the straw will be fed during cold weather.

## Testing

The OSU College of Veterinary Medicine has refined the laboratory procedures for measuring amounts of specific endophyte alkaloids in forages using high-performance liquid chromatography. Testing for ergovaline and lolitrem B is available to the public, at a cost of \$40 per sample for either test. The laboratory can analyze hay, straw, pellets, or grass plants from pastures.

It is important to submit a representative sample for testing. Take hay or straw samples with a core forage sampler. (Ideally, the core sampler should have an exterior diameter of ½ inch and a sample length of at least 12 inches.) Sample by centering the core sampler in the end of a bale and drilling horizontally. Take at least 1 core from each of 20 bales for each lot. Ways to take a representative sample might be sampling every fourth or fifth bale going around the stack or taking at least five random samples from each side of the stack.

To sample a pasture, take grass plants from at least 20 locations. Air dry the grass, not in direct sunlight. Seal samples in a polyethylene freezer bag and label them with your name, phone number, a sample identification, and the test you want performed (ergovaline or lolitrem B). Send samples, along with the payment, to the address below. The laboratory phone number is 541-737-2872.

Endophyte Service Laboratory 139 Oak Creek Building College of Agricultural Sciences Oregon State University Corvallis, OR 97331

This testing service is used by Oregon straw exporters, who test each 200-ton lot of grass seed straw shipped to Japan, Korea, or Taiwan. These countries require certification that ergovaline and lolitrem B levels are below the thresholds for producing animal disease.

The OSU Seed Laboratory, as well as private seed testing services, can test grass seeds for the presence of endophyte. The official sample and test required for issuing Endophtye Fungus Tested seed tags is provided only by the Oregon Department of Agriculture. Many seed growers obtain this certification if they plan to sell their seed out of state. An endophyte seed test result shows the percentage of seeds infected. For example, a 25 percent level means that 25 out of 100 seeds contain endophyte and will produce infected plants.

When the grass plant is dead, the endophyte also is dead. Therefore, it is necessary to test grass seed straw or screenings for the toxins rather than for the endophyte.

## **Novel endophytes**

In parts of the Southeast where heat and drought are severe, tall fescue pastures that are endophytefree do not persist as long as pastures planted with endophyte-infected tall fescue. To overcome this problem, researchers have looked for strains of the wild type endophyte that do not produce toxic levels of ergovaline, but that confer the other benefits of insect and disease resistance, drought tolerance, etc. on the grass. They have inserted these "novel" endophyte strains into tall fescue varieties. Some of the varieties have been patented and are currently marketed by seed companies. There are many strains of nontoxic endophyte, but only certain tall fescue varieties are a good match; that is, the endophyte can be maintained at a high enough level to help the variety perform better. Lengthy field trials are necessary to determine how a combination performs with respect to stress tolerance, and these trials are going on in several states.

In the cooler temperatures of the Pacific Northwest, tall fescue does not need the endophyte to perform well in pastures. A few Oregon grass seed growers are producing the seed of novel endophyte varieties for sale in the Southeast.

A number of the novel endophyte strains in both tall fescue and perennial ryegrass have been identified in other countries. Perennial ryegrass varieties with novel endophytes are currently being marketed by New Zealand seed companies.

### For more information

The Oregon Tall Fescue Commission has produced a free brochure, *Tall Fescue Endophyte Concepts*. Their telephone in Salem, Oregon, is 503-585-1157.

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