A Fungal Endophyte in Tall Fescue: Incidence and Dissemination

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ABSTRACT

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When tall fescue (*Festuca arundinacea*) is infected by the fungal endophyte *Epichloë typhina* (also referred to as *Acremonium coenophialum*) it causes summer toxicosis in grazing cattle. The endophyte was shown to be widely distributed and present at high levels of infestation in tall fescue fields in Kentucky. In individual plants the endophyte was found (in decreasing order of concentration) in leaf sheaths, seeds, crowns, stems, leaf blades, and roots. Very low amounts were recovered from leaf blades and roots, and high concentrations were found in the leaf sheaths and seeds. The endophyte was disseminated by seed and not by wind, rain, pollen, or mowing. During 4 yr the incidence of endophyte did not change significantly in experimental plots and fields managed for seed production (SP). Higher endophyte levels were found in SP plots than in those managed for hay-pasturage (HP). Hay yields and seed production in cultivar Kenhy tall fescue SP and HP plots were independent of the levels of the endophyte. The geographic origin of the endophyte in tall fescue, and the nature of the host-fungus relationships are discussed.

Additional key words: epidemiology, forage crops, mutualism.

Tall fescue (*Festuca arundinacea* Schreb.) is grown on 12–14 × 10^6 ha in the United States from Florida to Canada. It is the predominant cool-season pasture grass species grown in the transition zone between the latitudes of Indianapolis, IN, and Macon, GA, and the meridians of eastern Kansas and the eastern edge of the Appalachian piedmont area (5,6). The predominant cultivar grown in the transition zone is Kentucky 31 (Ky 31). Tall fescue has many desirable agronomic characteristics when used as cattle forage (5). However, animals grazing tall fescue often exhibit symptoms of fescue toxicosis. While several disorders are associated with fescue toxicosis, summer syndrome or summer toxicosis (7,13,14) is one of particular interest because this malady affects cattle grazing tall fescue that is infected with an endophytic fungus (2,8,16–18, 32). This fungus has been identified as *Sphacelia typhina* (Pers.) Sacc., the imperfect state of *Epichloë typhina* Fr. Tul. (2). Recently, Morgan-Jones and Gams (25) have discounted *Sphacelia* as the imperfect state of *E. typhina* and reclassified it and the endophyte of tall fescue as *Acremonium typhinum* Morgan-Jones & Gams and *Acremonium coenophialum* Morgan-Jones & Gams, respectively.

Symptoms of summer toxicosis include: increased respiration rates, elevated rectal temperature, extensive salivation, rough hair coat, reduced feed intake, decreased body weight gains, and reduced milk production. It has been estimated that fescue toxicosis costs livestock producers $50–200 million annually (9,12).

The fungus appears to exist as parasitic biotypes in many grasses (2). In certain species, such as tall fescue and perennial ryegrass (*Lolium perenne* L.), only symptomless intercellular systemic infections occur (2,28). In other grasses, such as bent grass (*Agrostis perennans*), prairie wedge grass (*Sphenopholis obtusata*), and orchard grass (*Dactylis glomerata*), symptoms of choke disease caused by *E. typhina* are produced and are characterized by destruction of the inflorescence (22,31,34). In New Zealand, the endophyte in perennial ryegrass has been reported to cause the neuromuscular disorder of sheep, known as ryegrass staggers (10,23).

Comparisons by enzyme-linked immunosorbent assay (ELISA) of isolates of the fungus from bent grass, tall fescue, and perennial ryegrass indicate that they contain similar antigens and therefore are closely related (11,19,21).

The endophyte is found in high amounts in tall fescue seed (20) and has also been detected in leaf sheaths, stems, and crowns but only rarely in leaf blades or roots (1).

Infected seed appears to be the only means of dissemination of the tall fescue endophyte in the field (2,29). The incidence and mode(s) of dissemination of the fungus is of primary epidemiological importance as it pertains to increases in the level of endophyte in existing and newly established pastures and to methods of control.

The objectives of this study were to determine the incidence and distribution of the fungus in the tall fescue plant, mode(s) and rate of dissemination, and the geographic origin of the endophyte. In a companion study (33) methods of control of the tall fescue endophyte were evaluated.

MATERIALS AND METHODS

Unless otherwise stated, detection of the endophyte in the seeds and dried vegetative portions of tall fescue plants was by ELISA, as previously described (21).

Distribution and incidence. Nine infected tall fescue plants of experimental line G1-307 were dug from the field in the winter; their roots were washed, and the plants were grown hydroponically in the greenhouse in aerated Hoagland's solution (15). The plants in three replicate groups of three plants each were harvested after 8 wk, and divided into root, crown, leaf sheath, leaf, blade, and stem tissues.

Samples for ELISA testing were dried at 60 C, ground, and placed in duplicate wells of an ELISA plate. The plate also contained ground fungus dilution standards (21). Concentrations of the endophyte in the various plant tissues and in seed collected from field-grown G1-307 plants were determined by comparison with the dilution standards.

The incidence of the endophyte in cultivar Ky 31 tall fescue

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pastures in Kentucky was determined in the spring of 1981 and 1982. The survey in 1981 consisted of randomly sampled mature panicles from 200 fields in 42 of the 120 Kentucky counties. In the spring of 1982 an in-depth survey was conducted in 37 pastures in Fulton, Trigg, Hardin, and Fayette counties. Integrated-pest-management scouts collected 30 samples of three to five culms per sample in the boot stage from a 0.2-ha area within each field. Within each of the four counties, stands were divided into three age groups, 2–5, 6–9, and 10 or more years old. The survey included a questionnaire to determine farmer recognition of the symptoms of summer toxicosis.

In an attempt to determine original sources of endophyte in commercial fields, the presence of the endophyte was tested in tall fescue seed or stem tissue from the Suiter farm (established prior to 1890 and the origin of Ky 31 cultivar), the McCauley farm (a 20.2-ha source of Ky 31 certified seed planted about 1948–1950), from Poland (13 ecotypes), and from France (four ecotypes).

Tall fescue plots of cultivars and experimental strains established by broadcast seeding (15–17 kg of seed per hectare) in 1974 and 1979 for agronomic research were tested to determine annual changes in incidence of the endophyte. The plots were managed for HP or for SP. When managed for HP, the plots were clipped for hay and then mowed approximately every 28 days thereafter to simulate pasture conditions. When managed for SP the aftermath was removed immediately after seed harvest and again during late August after which the grass accumulated until the first autumn freeze when it was harvested as stockpiled growth. The HP plots were fertilized with N at 35 kg/ha on approximately 1 March, 1 June, and 1 September of each year while the SP plots were fertilized with N at 69 kg/ha around 1 September and 1 December of each year.

In the first experiment, G1-307 tall fescue (0.81 ha) was established in the fall of 1979 and managed for SP. The percent infected seed was determined in the original seed source as well as from harvested seed for each successive year through 1983. Harvested seed were collected, cleaned, and placed in 22.7-kg bags. Bags (about eight per year) were sampled (25 gm/bag) with a grain trier. The subsamples were combined, and 150 seeds were individually tested for the endophyte.

In the second experiment, 13 named cultivars and experimental lines of tall fescue and perennial ryegrass were established in the fall of 1979 in 4.6 × 22.9 m plots as a randomized split block design with each entry replicated three times. Entries were superimposed as subplots on main plots that consisted of the two management systems. The percent infestation of each SP replicate was determined by analysis of seed harvested in 1980, 1982 and 1983 from the five cultivar Kenhy entries. Floral tillers were harvested from the HP and SP plots in the spring of 1983 and tested for the endophyte. Endophyte analysis involved sampling first and second internodal stem tissue and immature seed.

The entries were harvested each year (1980–1983), and hay yields, aftermath yields, fall accumulation growth, and seed yields were determined. The vegetative matter and seed were harvested in 27 × 20.1-m portions from each plot. After harvest, the remaining growth was removed by mowing at right angles to the direction the samples were taken for yield. The entries were also scored for fall stand and maturity at hay harvest.

Dissemination and transmission. In a third experiment, G1-307 and Ky 31 were established in 1974 in adjacent 1.42-ha fields having a 203-m boundary in common. The G1-307 seed was >98% infected (current year seed) while the Ky 31 was planted from 2-yr-old seeds in which the endophyte was no longer viable. The plots were managed for HP until 1982 except in 1979 when seed was produced from the G1-307 tall fescue. In the spring of 1982, the Ky 31 plot was surveyed for the endophyte by collecting basal stem tissue on transect lines perpendicular to the boundary line, with samples being collected 0, 4.57, 9.15, and 13.72 m from the boundary and then at 9.15-m intervals until the opposite boundary line was reached.

To determine if animals can disseminate infected seed, 5.5 kg of Ky 31 seed (90% germination, 63% seedling infection) was given to a 318-kg steer via a rumen cannula over a 3-day period. The animal was also fed an oral fescue-corn diet. A portion of the feces (~3 g) collected on the third day was spread on the surface of a Pro-Mix, sand, soil mixture (1:1:1, v/v) in 7.6-cm-diameter pots. As a control, fresh feces from a steer not consuming seed was mixed with the Ky 31 seed, allowed to incubate at room temperature for 0, 24, and 48 hr and then spread on the soil surface. After 10 wk of growth, stem samples from fescue plants were tested for the endophyte. Seed (Ky 31) was also separated from the third-day feces collection by washing it on a wire screen. Whole seed (400) were removed, and the percent germination was determined on filter paper.

Reciprocal crosses of infected and noninfected parents were made using controlled pollinations to investigate the mode of transfer of the endophyte from parent to progeny. Kenhy tall fescue plants previously identified as being endophyte-infected or non-infected based on microscopic examination of culm pith tissue (29) were crossed in all combinations. Endophyte in the vegetative tissue and seeds was determined by GLC analysis for pyrrolizidine (*N*-formyl and *N*-acetyl loline) alkaloids (8). The accumulation of these alkaloids has been shown to be associated with endophyte infection (8).

Attempts were made to artificially infect tall fescue plants to obtain well established endophyte-infected plants. Isolations of the fungus were made from endophyte-infected tall fescue tissues 1–3 mo previously. These were cultured in a nutrient solution (M 43) (2) for 3 wk on a reciprocal shaker followed by 2–3 wk standing, or on cornmeal-soytone agar (21).

Twenty-six G1-307 tall fescue plants were inoculated. Twenty were flowering and were free of the endophyte. Flowering was induced by placement in a sand-filled, plastic-lined trench in the lathhouse from September 1981 to February 1982. The remainder were nonflowering endophyte-free tall fescue plants that had been maintained in the greenhouse at 20–25 C. Stems from all plants were sampled immediately before inoculation, and were tested for endophyte infections.

Plants were inoculated using one of the following methods: (i) injection of mycelial fragments and conidia into stem and crown tissue using a hypodermic needle, (ii) spraying a suspension of mycelial fragments and conidia, and (iii) application of agar disks containing the fungus. In methods ii and iii, inoculum was applied to freshly cut surfaces of stem tissue. Plants were misted with distilled water and enclosed in plastic for 60–72 hr following inoculation. Stems from the inoculated and uninoculated, endophyte-free control plants were harvested ~6 mo postinoculation, and tested for the endophyte.

RESULTS

Distribution in the plant. The tall fescue endophyte was detected in vegetative tissue and in seed, although the amounts present varied widely (Table 1). The lowest concentration was in the roots and may have represented contamination, because the amount reported was found in only one of the three replicate root samples. The highest concentration of fungus was in the leaf sheaths. The amount found in the seeds nearly equaled that in leaf sheaths. Both stem and crown tissue also contained substantial concentrations of fungus.

Incidence. A survey in 1981 of 200 fields from 42 counties in Kentucky indicated that 97% of the fields were infested. The 1982 survey of 37 fields indicated that all fields were infested and that regardless of age of the field, a substantial number had high levels of infestation (Table 2). The average percent infestation for all age groups was 64%. Grouping the number of infested fields, by percent range, indicated that most were in the 67–100% range and there was no correlation in age of the stand and incidence. None of the symptoms of summer toxicosis listed in the questionnaire were recognized by the respondents.

Seven of the ecotypes (land cultivars) collected from Poland were infested, as well as one of four ecotypes from France. The endophyte was found in 26 of 28 samples from the Suiter farm and 29 of the 30 samples from the McCauley farm.

The 1982 survey (Table 2) indicated that four older fields (10 or

more years) were in the 10–33% infested range. This suggested that the endophyte may not rapidly increase over long periods of time and hence is limited to seed dissemination. In order to further test this hypothesis but within a shorter time period, plots that were managed for SP were analyzed to determine infestation levels. In the first experiment, involving a G1-307 tall fescue field planted in 1979, there were no significant differences in endophyte levels in seeds during the four succeeding years (Table 3).

In a second experiment involving 4.6 × 22.9 M randomized split plots of cultivars and experimental lines of tall fescue managed for SP, percent infection in 1982 ranged from 7 to 90% (Table 4). In the five Kenhy entries, the level of endophyte infection among the four years was not significantly different. Since these split plots were managed also for HP, it was possible to determine the effect of the two management practices on the presence of the endophyte in the five Kenhy entries (Table 5). The percent infection values from the SP plots were not significantly different when immature panicles sampled in the spring of 1983 (Table 5) were compared to seed harvested in the summer of 1983 (Table 4). However, cultivar management practice significantly affected levels of endophyte in 1983. Panicles from HP plots showed lower mean percent infection than panicles from SP plots.

The effect of endophyte infection on growth and reproduction of the five Kenhy entries managed for SP or HP was determined (Table 6). There was no significant difference between SP plots among the four years and the HP plots in 1983. However, there was a significant difference between certain entries for mean hay and aftermath yields for 1980–1983 from the HP plots. Correlation analysis for this harvest period indicated that the differences were independent of endophyte levels (determined in 1983) among entries. Aftermath yields in 1983 were much lower than in previous years because of drought in July–September.

Dissemination and transmission. Increases in endophyte levels can occur in fields which border a highly infested field and are managed differently. This appeared to be the case in the Ky 31 and G1-307 tall fescue fields planted in 1974 that shared a common border. Analysis of data from these fields in 1982 indicated that the G1-307 and Ky 31 fields were >98 and 8.5% infested, respectively. However, 86% of the infestation (18 of 21 positive samples) in the Ky 31 field was localized within 13.7 m from the Ky 31 border and G1-307. In 1979, the G1-307 field was managed for SP. The seed and aftermath harvesting operations involved movement of equipment and materials across the common border, and it appears that the infected plants in the Ky 31 field may have originated from seed shatter during harvest.

Animal feeding on infected tall fescue that is producing seed may disseminate infected seed via feces into pastures. This hypothesis was tested by collecting feces from a steer fed infected seed via a rumen cannula. Numerous whole seed were recovered in the feces. This seed germinated (50% of the nonfed controls) and produced infected seedlings (40% of the nonfed controls).

Analysis of tall fescue forage and seed for pyrrolizidine alkaloids, used as an indirect measurement of infection, clearly indicated that the endophyte was only maternally transmitted and not pollen transmitted (Table 7). Crosses between noninfected maternal parents and infected paternal parents produced noninfected progeny. Infected progeny occurred only when the maternal parent was infected.

Attempts to infect endophyte-free plants by inoculating stem and crown tissue and cut stem tissue were unsuccessful. None of the inoculated tall fescue plants contained antigens of the endophyte 6 mo postinoculation.

TABLE 1. Distribution and amount of the fungal endophyte in experimental line G1-307 tall fescue plants and seeds[w]

Plant portion	Endophyte concentration (mg/g dry wt tissue)[x] Mean
Leaf sheath[y]	4.60 (± 0.24) a
Crown	1.15 (± 0.33) b
Stem	1.04 (± 0.16) b
Leaf blade	0.14 (± 0.09) c
Roots	0.01 (± 0.02) c
Seed[z]	2.20 (± 1.58)

[w] G1-307 tall fescue is an experimental strain derived from perennial (*Lolium perenne*) and annual ryegrass (*Lolium multiflorum*, Lam.) × tall fescue hybrids.

[x] Mean values in column, standard deviations in parentheses, followed by the same letter are not significantly different ($P = 0.05$) according to Duncan's new multiple range test performed on log-transformed data.

[y] Infected field plants harvested after an additional 8 wk of growth in nutrient solution in a greenhouse and divided into three replicates of three plants each.

[z] Seed harvested from field-grown plants (>98% infected) and 30 individual seed tested for the endophyte. Range = 1.1–6.8.

DISCUSSION

Endophyte-infected tall fescue is seed disseminated and appears to be widely distributed in Kentucky and the transition zone. In the 1982 survey in Kentucky, the number of fields with high levels (>66%) of infestation was sufficient to indicate that summer toxicosis should be a major problem. While both the threshold level of infestation and/or level of the unidentified toxic chemical entity(s) responsible for summer toxicosis are unknown, animals grazing tall fescue with 57% incidence of infection had major toxicosis symptoms (*unpublished*). On the other hand, animals fed fescue with a 44% level of infection under controlled temperature conditions showed reduced summer toxicosis symptoms (18).

Our studies indicate the endophyte is not disseminated by pollen, wind, or rain. In addition, the incidence experiments (Tables 3 and 4) suggest that mowing also does not disseminate the fungus. The common management practice in these experiments was to mow infected and noninfected plants within plots and across plots that had different levels of infestation.

We have been unable to infect tall fescue plants by inoculating them with endophyte grown in culture, although Latch and Christensen (24) have recently infected young seedlings of both perennial ryegrass and tall fescue with their respective endophytes.

The potential for infection of tall fescue (via the flower stigmas) by conidia and ascospores from grasses exhibiting symptoms of the choke disease is unknown. Serological studies indicate a close relationship between isolates of the tall fescue and perennial ryegrass endophytes, and the choke disease pathogen *E. typhina* (11,19,21).

While the level of viable endophyte in a seed lot may be one of the primary factors in determining the amount of infestation in newly planted pastures, the management system may also affect endophyte levels. Infestation levels did not change over 4 yr in the

TABLE 2. The 1982 survey for incidence of the tall fescue fungal endophyte in 37 pastures of different ages in four counties in Kentucky[a]

Field age (yr)	Fields (no.)	Infected plants (%) Avg.	Range	Fields (no.) infested in indicated range 10–33%	34–66%	67–100%
2–5	12	60	18–100	4	3	5
6–9	11	72	27–97	2	0	9
10+	14	59	10–100	4	3	7
Grand avg.		64				
Total no. (%) of fields				10 (27%)	6 (16%)	21 (57%)

[a] Thirty stem samples (boot stage) from 0.2 ha per field were analyzed for the endophyte.

two experimental SP plots. However, when endophyte analysis was made in 1983 of flower panicles from HP and SP plots established in 1979, a trend towards lower infestation levels was detected in the HP plots. The reason for this trend may be differences in fertilization, mowing practices, and/or some other unknown factor. It was not possible to determine if this observed trend resulted from consistent year to year changes in endophyte level or from a change in one particular year.

Of primary importance in understanding why the endophyte is widely disseminated at varying levels of infestation is a knowledge of how seed increases occurred during the establishment of Ky 31 and the viability of the endophyte in stored seed. E. N. Fergus, the discoverer of Ky 31, reports (*personal communication*) that seed harvested in the spring from the Suiter farm and various experimental plots between 1931 and 1943 was planted with minimum delay in the fall. With the release of Ky 31 in 1943 and its subsequent rapid acceptance by farmers there was again almost total planting of current year's seed production. The high infestation on the McCauley farm, a source of Ky 31 certified seed in the late 1940s and early 1950s, indicates one source of highly infected seed. It is known that endophyte viability in tall fescue and perennial ryegrass seed is temperature dependent and decreases or is lost during storage for 1–4 yr (3,20,23,29). For example, a total loss of endophyte viability occurred after 7–11 mo of storage at 21 C, but no loss occurred after 19 mo of storage at 6 C (33). Since most storage conditions involve variable temperatures, it appears that 6- to 12- mo-old seed could still contain substantial amounts of viable endophyte.

The lack of change in infestation levels, the number of infected ecotypes collected from Europe, and the high infestation level found on the Suiter farm support the hypothesis that the current levels of infestation in the transition zone are directly related to the European introduction of infected tall fescue into the U.S. during the last half of the 19th century and seed dissemination of the endophyte.

We suggest that a unique interaction exists between host grasses (perennial ryegrass and tall fescue) and the endophytic fungus. While the exact nature of the interaction has not been determined, we consider this relationship to be mutualistic or (at least in tall fescue) commensal in which the plant is neither helped nor harmed by the fungus. It is clear that the endophyte receives an advantage by having its reproduction integrated into that of the host(s). While the fungus in tall fescue has no apparent effect on growth and reproduction of the plants, infection does cause the production of pyrrolizidine alkaloids (8). Levels as high as 0.5% by dry weight have been found in infected tall fescue plants (18). The role and function of the alkaloids in infection, plant growth, or summer toxicosis is unknown. The *N*-formyl and *N*-acetyl loline alkaloids have not been found in perennial ryegrass (*unpublished*).

The possible benefits to host plants are related to the effect of the infection on insect and animal behavior. With reduced feeding by herbivores, especially during hot weather, there is less tendency for endophyte-infected tall fescue pastures to be overgrazed. Rodents (rats and mice) also consume considerably less infected tall fescue seed as compared to noninfected seed (27, and *unpublished*).

The most striking phenomenon involving any selective advantage of infected over noninfected plants is that of enhanced insect resistance. Ryegrass cultivars with high levels of endophyte have been reported to be resistant to sod-web worm (*Pediasia trisecta*), Argentine stem weevil (*Listronotus bonariensis*), and bluegrass billbug (*Sphenophorus parvulus*) (11,26,30). Insect resistance has not been reported in field-grown infected tall fescue. However, in laboratory experiments, feeding by the Argentine stem weevil (4) and oat bird cherry aphid (*Rhopalosiphum padi*) (*unpublished*) was reduced when the insects were confined on endophyte-infected tall fescue plants compared to those similarly confined on endophyte-free plants.

In nature, endophyte-infected plants may have a selective advantage over those that are endophyte free, but the tall fescue endophyte now may be at a disadvantage because of man-made selection pressure for noninfected plants. This has arisen because the endophyte has only one apparent mode of dissemination which is susceptible to treatments that reduce or destroy fungal viability. This aspect of endophyte control, as well as other methods of control, is the subject of a subsequent publication (33).

TABLE 3. Incidence of the endophyte in seed of experimental line G1-307 tall fescue during 4 yr of management for seed production[a]

Year	Infection (%)[b]	Range (%)[c]
1979	51	37–70
1980	58	38–77
1981	50	37–70
1982	57	40–73
1983	45	37–50

[a] The 0.81-ha stand was planted in fall of 1979, and seed were harvested in July of each succeeding year.
[b] Analysis of five replicates of 30 individual seed each.
[c] Analysis of variance of arc sine square root-transformed data indicated no significant ($P = 0.05$) differences in endophyte levels between the years.

TABLE 4. Incidence of the fungal endophyte in cultivars and experimental lines of tall fescue and perennial ryegrass after 4 yr of seed-production management[a]

Cultivar or line	Infection (%)[b,c]		
	1980	1982	1983
Ky 31		10	
MO 96		33	
New Zealand		29	
Grimalda ryegrass		33	
SYN 470[d]		80	
SYN 474 and 480		90	
SYN 479		77	
SYN 471		72	
Kenhy breeders (KY-1)	72 (60–83)[e]	74 (67–80)	73 (64–81)
Kenhy certified (OR-2)	11 (7–17)	7 (3–10)	12 (5–15)
Kenhy certified (OR-3)	22 (13–27)	28 (23–33)	26 (10–33)
Kenhy certified (KY-4)	8 (7–13)	8 (0–17)	7 (3–10)
Kenhy certified (OR-5)	48 (40–50)	52 (50–55)	57 (50–57)

[a] Overall split-plot design included hay-pasture and seed production management.
[b] Percent infection for all cultivars except Kenhy entries determined on 30 individual seed, 10 per replicate. For five Kenhy entries, 90 individual seed, 30 per replicate.
[c] Analysis of variance of data transformed by arc sine square root indicated no significant difference in endophyte levels for the Kenhy entries among the years.
[d] Original planting in 1979 contained six synthetic experimental strains. In 1980 and 1981, two of these strains went out of stand.
[e] Figures in parentheses show range of infection among the replications.

TABLE 5. Effect of hay-pasturage and seed production management on the presence of the fungal endophyte in cultivar Kenhy tall fescue entries[a]

Entry	Type of management[b]	Infection (%)[c,d]	Range (%)[e]
Kenhy breeders (KY-1)	SP	76	60–87
	HP	66	59–74
Kenhy certified (OR-2)	SP	10	7–13
	HP	2	0–4
Kenhy certified (OR-3)	SP	33	27–37
	HP	18	13–20
Kenhy certified (KY-4)	SP	7	3–10
	HP	1	0–3
Kenhy certified (OR-5)	SP	57	47–63
	HP	42	33–53

[a] From the same split-plot experiment as in Table 4.
[b] SP, seed production; HP, hay-pasturage.
[c] Percent infection determined in 1983 by an analysis of 30 panicles collected from each of three SP and HP replicates.
[d] Analysis of variance of arc sine square root transformed data indicated a significant ($P = 0.05$) effect of entry and type of management practices on the levels of endophyte.
[e] Range of infection among the replications.

TABLE 6. Effect of infection by the fungal endophyte on hay-pasturage and seed production in five cultivar Kenhy tall fescue entries[a]

Type of production	Means of the years of study (1980–1983)						
	Ken-1	Ken-2	Ken-3	Ken-4	Ken-5	LSD ($P = 0.05$)	C.V. (%)
Hay-pasturage management							
Infection[b] (%)	66	10	13	7	42		
Hay yields (DM[c] kg/ha)	2,780 (2,645)[d]	2,735 (2,623)	2,892 (2,713)	2,579 (2,579)	3,228 (2,959)	291 (NS)[c]	12.5
Aftermath yields (DM kg/ha)	2,309 (1,252)	2,152 (1,054)	2,130 (1,009)	2,107 (1,121)	2,242 (1,121)	135 (NS)	7.4
Fall stand estimates (%)	88 (83)	80 (72)	81 (72)	87 (82)	88 (83)	NS (NS)	12.4
Maturity at harvest[e]	7.0 (9.0)	7.5 (8.8)	7.2 (8.2)	8.0 (9.2)	8.0 (9.4)	NS (NS)	17.4
Seed production management							
Infection[f] (%)	75	12	26	7	57		
Fall accumulated growth (DM kg/ha)	1,390	1,255	1,300	1,300	1,278	NS	15.0
Seed yield (kg/ha)	330	342	325	341	404	NS	21.6
Fall stand estimate (%)	86	81	83	85	85	NS	5.7

[a] Five Kenhy entries same as those in Tables 4 and 5.
[b] Percent infection 1983 (flower panicles).
[c] DM = dry matter; NS = not significant.
[d] Numbers and letters in parentheses are 1983 yields.
[e] Maturity classes: 1 = vegetative, 3 = early bloom, 5 = late boot, 7 = early head, 11 = early bloom, and 13 = full bloom.
[f] Percent infection 1983 (seed).

TABLE 7. *N*-formyl and *N*-acetyl alkaloid (FALA) content of forage, seed, and progeny of reciprocally crossed fungal endophyte infected and noninfected tall fescue parents

	FALA (μg/g tissue)					
	Cross (females × males)[a,b]			Reciprocal crosses (females × males)		
	A × X	B × Y	C × Z	X × A	Y × B	Z × C
Maternal parent's forage	910	1,442	1,432	T[c]	T	T
Maternal parent's seed	2,475	3,384	5,593	19	148	T
Progeny seedlings	1,107	1,482	1,363	0	0	0

[a] Average of three replications.
[b] Endophyte status, determined by staining, indicated that parents A, B, and C were endophyte positive; parents X, Y, and Z were endophyte negative.
[c] T = trace.

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