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## Pyrrrolizidine alkaloid accumulation in tall fescue during plant growth and development

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**Key words :** lolines

**Introduction** Tall fescue, *Lolium arundinaceum* (Schreb.) Darbyshire, infected with the endophytic fungus, *Neotyphodium coenophialum* (Morgan-Jones and Gams) Glenn, Bacon, and Hanlin, accumulates peramine, ergot and loline alkaloids. The loline alkaloids are present in much greater amounts than the other alkaloids. In the common endophyte strain the predominant pyrrrolizidine alkaloids are N-formylloline (NFL) and N-acetylloline (NAL). Accumulation of these alkaloids is a significant determinant for ecological fitness. The objectives of this study were to determine development of *N. coenophialum* in tall fescue and the accumulation of NAL and NFL in different plant tissues and clippings.

**Materials and methods** Flowering plants were collected from field grown plants at Lexington, KY in 1982 and 1983 (Burhan, 1984). Plants were grown in pots in the glasshouse with 4 replications in a split-plot design with time and frequency of harvest as sub-plots. Harvests were made weekly from pots that had not been previously harvested and regrowth tissue was harvested at 3 week intervals. NFL and NAL was measured by gc/FID. Endophyte was measured by ELISA of Johnson et al. (1982).

**Results and discussion** In flowering plants greatest loline abundance was found in the spikelet and much lesser amounts in descending order in rachis, stem, leaf sheath and leaf blade. Endophyte concentration was greatest in the rachis. Leaf blades contained the lowest concentration of NFL, NAL and endophyte in these flowering plants. Samples were taken at anthesis and most likely levels of alkaloids and endophyte would have increased as the spikelet matured. In flowering plants the amount of NAL + NFL in the stem, rachis and leaf blade had a positive and significant linear correlation with the amount of endophyte in the respective tissue. Small amounts of NFL and NAL were measured in roots from plants grown in soil or sand culture but not in roots grown in solution culture. Presence of these alkaloids in all tissues examined, strongly suggests translocation within the plant. Koulman et al. (2007) measured NFL in sap from cut leaves and guttation water of tall fescue. The observation that the lolines are in the guttation water and thus found on the surface of the leaves may be significant for insect deterrence.

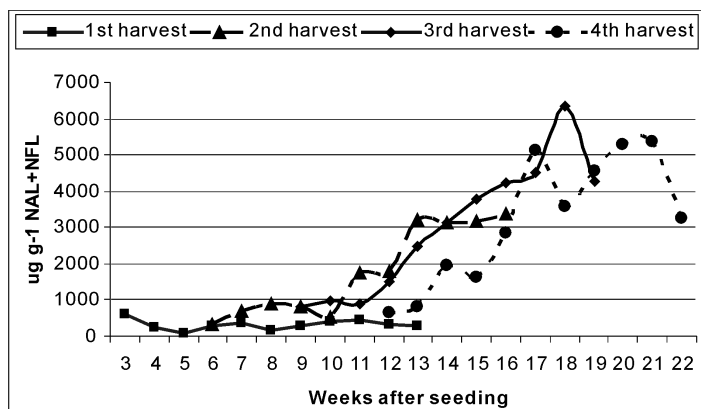


Figure 1 Harvest frequency effect on alkaloid accumulation.

In vegetative plants the greatest amount of lolines are found in the pseudostem with lesser amounts in the leaf blade. Pseudostem tissue also contained as much as two orders of magnitude more endophyte than the blade ( $\sim 1000$  vs  $10 \text{ g g}^{-1}$ ). However, there was no significant correlation between endophyte and NAL and NFL concentration. This result is probably because of translocation of the alkaloids and the dynamic of endophyte growth and accumulation of alkaloids. NFL + NAL concentrations were very similar in initial harvest from 3 to 13 weeks after seeding (Figure 1). However, with additional harvests at 3 week intervals the alkaloid content was much higher. Plant maturity alone was not sufficient to cause an increased alkaloid concentration as indicated by similar alkaloid accumulation at the initial

harvest time with 10 weeks difference in age. The increase in alkaloids with clipping may be result of mobilization of plant reserves for regrowth and thus also available for endophyte biosynthesis of the alkaloid.

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