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Combined analyses of steer performance from independent tall fescue grazing trials

Robert Wayne Thompson

To the Graduate Council:

I am submitting herewith a dissertation written by Robert Wayne Thompson entitled "Combined analyses of steer performance from independent tall fescue grazing trials." I have examined the final electronic copy of this dissertation for form and content and recommend that it be accepted in partial fulfillment of the requirements for the degree of Doctor of Philosophy, with a major in Plant, Soil and Environmental Sciences.

Henry A. Fribourg, Major Professor

We have read this dissertation and recommend its acceptance:

John H. Reynolds, William L. Sanders, John C. Waller, J.B. McLaren

Accepted for the Council:

Carolyn R. Hodges

Vice Provost and Dean of the Graduate School

(Original signatures are on file with official student records.)

To the Graduate Council:

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Combined Analyses of Steer Performance from Independent

Tall Fescue Grazing Trials

A Dissertation

Presented for the

Doctor of Philosophy

Degree

The University of Tennessee, Knoxville

Robert Wayne Thompson

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DEDICATION

This dissertation is dedicated to my mother, Betty Christine Kacey Johnston. Thank you Mom for providing me with the emotional and financial support I needed to complete my PhD training.

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ABSTRACT

Most of the information concerning the effects of the tall fescue (*Festuca arundinacea* Shreb.) endophytic fungus (*Acremonium coenophialum* Morgan-Jones Gams) on beef (*Bos taurus* L.) steer performance has been obtained from grazing trials conducted as independent endeavors. These trials may be related over space and time. Datasets from 12 trials conducted during the last 13 years at nine locations in seven eastern US states were pooled to provide combined estimates of steer daily gains on tall fescue pastures free (E-) of or infested (E+) with *A. coenophialum*. Treatments included E- fescue ($\leq$ 5% E+); moderately infested fescue ($\geq$ 20% to $\leq$ 35% E+); highly infested ($\geq$ 50% to $\leq$ 97% E+); and in tall fescue-clover (*Trifolium* spp. L.) mixtures, endophyte-free (**E- CL**), moderately infested (**MECL**), and highly infested (**HECL**) at the same E+ levels with about 25% and 10% clover in spring and summer stands, respectively. Spring, summer, spring plus summer together, and fall plus winter together datasets were analyzed separately using Henderson's mixed model procedure (MMP). In addition to incorporating the variance components of the random effects into the mixed model equations, mean daily gain estimates were adjusted for the initial weights and steer grazing days ha^{-1} covariates. Seasonal steer performance generally reflected pasture E+ level and clover incidence. Mean daily gains were variable for the

treatment X location combinations, but most of the variation occurred within highly infested treatments. Mean daily gains were comparable for models analyzed with and without the steer grazing days ha^{-1} covariate, but the standard errors of the means were smaller for those models which included this covariate. The MMP permitted the estimation of the fixed effects of treatments and treatments X locations over a broad inference space of future years and different pastures. Since the combined analysis was able to estimate treatment effects which were not obtainable in each discrete study, combining datasets may be a feasible way to circumvent some of the financial and logistical constraints that force undesirable comprises in the conduct of grazing and other expensive or time-consuming research. The establishment of cooperative projects, using common treatments and identical protocols, would further increase the sensitivity of combined analyses.

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CHAPTER 1

LITERATURE REVIEW

Developments in Combined Analyses

Methodology for combining experiments from repeated research studies has a long history. Early examples of combining data are found in replicated astronomical and physical measurements. In the first half of this century, modern statistical methods began to be developed for the analysis of discrete agricultural experiments. Since agricultural experiments in particular lend themselves to replication, this led to the development of statistical techniques for merging raw data or results.

Two central aspects of combining experiments were rapidly recognized (Hedges and Olkin, 1985). One involved methods for collecting the body of information to be summarized. This opened the door to a multiplicity of problems and questions, such as the steps that should be taken to guarantee objectivity, and whether some studies should be excluded because of inadequacies in design or execution. In developing an understanding of these kinds of problems and questions, it is often helpful in reviewing the studies to summarize the methodology and findings of each study.

The second aspect of combining independent studies assumes as a starting point that one has available a set of reasonably well-designed studies that address the same question(s) using similar responses, and which focus on the methodology needed for summarizing data (Hedges and Olkin, 1985). Since classical statistics address primarily the analysis of individual experiments, new formulations, models, and methods are usually required for a combined analysis.

Two distinctly different directions have been taken for combining evidence from discrete studies in agriculture almost from the very beginning of statistical analysis of agricultural data. One approach relies on testing for statistical significance of combined results across studies, and the other relies on estimating treatment effects across studies from raw data. Both methods date from as early as the 1930's or earlier, and continue to stimulate interest within the statistical research community to the present day.

Testing for the statistical significance of combined results from agricultural experiments is perhaps the older of the two traditions. Meta-analysis is the rubric used to describe quantitative methods for combining evidence across studies (Hedges and Olkin, 1985). Since meta-analysis usually relies on 'data' in the form of summary statistics derived from the primary analyses of studies, it is an analysis of the results of statistical analyses. These tests of the significance are sometimes called omnibus or nonparametric tests because they do not depend on the type of data or the statistical distribution of those data (Hunter et al., 1982).

One of the first proposals for a test of statistical significance of combined results (now called testing the minimum p or Tippett method) was given by Tippett (1952). Soon afterwards, Fisher (1936) proposed another method for combining statistical significance, or p-values, across studies. Pearson (1933) derived independently the same method shortly thereafter, and the methods variously called Fisher's method or Pearson's method were established. Research on tests of significance of combined results has increased dramatically since that time, and now well over 100 papers in the statistical literature have been devoted to such tests (Hedges and Olkin, 1985).

Although omnibus tests or tests of statistical significance are appealing because they can be applied universally and do not require that raw data be concatenated, they suffer from an inability to provide estimates of the magnitude of the effects being considered (Hedges and Olkin, 1985). Therefore, these tests appear to be of limited utility for combining results from grazing research.

In order to determine the magnitude of the effect of an agricultural treatment, a second approach was developed that involved combining numerical estimates of treatment effects. One of the early papers on the subject (Cochran, 1937) appeared shortly after the first papers on omnibus procedures. In this paper, Cochran defined the relative efficiencies of four types of means, the tests of significance for the mean response and for the variation in response from one location to another, and the estimation of the mean response when it varies from location to location. Additional work in this tradition appeared shortly thereafter (Yates and Cochran, 1938; Cochran, 1943). These researchers recognized some of the problems of combining numerical estimates of treatment, location, and year effects that we face today. In particular, they recognized that not all studies provide equally good data, and that the estimates of experimental error and the quality of the data reported with each study are not to be trusted completely.

The Ideal Conditions for Combining Datasets

Before discussing specific statistical procedures for combining data, it is helpful to consider what can be expected of a combined statistical analysis in the best possible situation. Perhaps the simplest example is one in which the raw data from several experiments are available and can be pooled directly. For example, suppose we have a series of **k** grazing trials, each of which is designed to investigate the effect of a treatment using an experimental/control group design. Assume that each trial measures the same response variable using the same management procedures so that the within-group population variances of the responses are similar. For convenience, one can arbitrarily fix the common within-group variances to be unity, although this is not essential. This situation is one in which the raw data from all grazing trials are directly comparable. In this idealized case, the assumptions of the analysis of variance will be met. The assumptions required are that the errors are normally and independently distributed and have equal variances for each treatment in the **k**th trial (Stroup et al., 1985). Diagnostic tests for normality and equality of variance should be standard procedures for all datasets. Examples of these tests using data from grazing trials are provided by Stroup et al. (1985).

What does one learn from the combined analysis of variance? The F-test for the main effect of trials tests whether the average value of the response (averaged over both experimental and control groups) differs across individual trials. Two other more important F-tests exist. The F-test for the treatment effect tests whether the treatment group performs differently than the control group, on the average, across all **k** grazing trials. The other F-test for the treatment by trials source of variation tests whether the treatment effect is consistent across trials. The interpretation of the statistical analysis is largely dependent on the last two F-tests. A large treatment effect with a negligible interaction indicates that the treatment produces a consistent effect across all trials. Even if the interaction is negligible, this fact cannot be taken as indicating no variation in the treatment differences from trial to trial, but only that such variation is likely to be smaller than can be determined by the arguments of fiducial probability (Yates and Cochran, 1938).

If the treatment by trial interaction is determined to be large, then the interpretations become more complicated. A significant interaction suggests that the treatment effect is larger in some studies than in others. Statements about the main effects must be qualified by the fact that treatment effects vary significantly across trials.

A significant interaction indicates that one should look for causes of variations in treatment effects across studies. Variations across trials in treatment, experimental procedure, conditions of measurement, or sample composition might help in the explanation of variations in treatment effect (Hunter et al., 1982). If a suitable explanatory variable is found, it should be included in the analysis as a blocking factor or covariate. The new analysis would reveal whether the new variable(s) accounted for a significant amount of variation in treatment effects and whether variations in the treatment effect across studies within levels of the new variable(s) remained significant.

One possible way to remove some of the residual variability among data from the combined studies is to use explanatory variables in an analysis of covariance. Covariance analysis can be used to increase the precision of comparisons among treatments in respect of the response **Y** by adjusting for the inequalities of the covariate **X** (Finney, 1989b). Covariance analysis is an often under-used procedure in agronomy and other disciplines. Possible climatic covariates include growing degree days, drought days, evapotranspiration rates, daily air temperatures and precipitation, *etcetera*. Possible animal covariates include initial weights, stocking rates or densities, frame size, age, *etcetera*. Possible soil covariates include base saturation or pH, water holding capacity, depth of A horizon, % of soil separates, *etcetera*.

Therefore, in the best possible case, where data from all studies can be combined directly, the combined analysis of variance has several features:

- The average trial effect can be estimated and tested across all treatments.
- The average treatment effect can be estimated and tested across all grazing trials.
- The consistency of treatment or trial effects can also be tested by the treatment X trial source of variation.
- The effect of explanatory variables that define differences among trials can be tested.
- The relationship between the response variable and the explanatory variable can be tested to determine if it is consistent among treatments.

The incorporation of other effects, such as years, into the analytical model(s) further complicates the combined analysis of variance. On the other hand, the principles for making valid tests of hypotheses remain the same as those of a combined analysis which considers only the effects of trials and treatments during a given grazing year.

Securing a Set of Random Locations and Years

It is usually impossible to secure a set of locations and years selected entirely at random. An attempt should be made to insure that the locations and possibly the spectrum of years actually used are a representative selection, but averages of the responses from such an assembly cannot be accepted with the same assurance as would the averages from a random sample (Yates and Cochran, 1938). On the other hand, comparisons between the responses at different locations and/or years are not influenced by lack of randomization in the selection of locations and years, except that an estimate of the variance of the response is required. The lack of randomization is then only harmful insofar as it results in the possible exclusion of locations and years of certain types and, in consequence, the range of conditions for treatment evaluation is narrowed (Yates and Cochran, 1938).

It seems appropriate to regard all effects of a combined analysis in both space and time as random variables except the general mean and the true effects of the treatments. This seems suitable because if one could tabulate all the values of, *e.g.*, the treatment X location interaction in the population, they would follow some frequency distribution from which the values in the data are a sample (Fisher, 1936). If the number of locations and/or years is reasonably large and these main effects represent a sample from some underlying distribution, then locations and/or years should be defined as random effects. Both locations and years can be considered as broad types of replications, with locations being replications or samples of the area for which information is desired, and years being replications or samples for future years.

Variance Heterogeneity

Much of the literature on combined analyses is for agricultural experiments that had identical treatments conducted over a random set of locations over the same years, or for the same location for several years, *e.g.*, performing a combined analysis on the yields of 10 cultivars of wheat (*Triticum aestivum* L.) at 7 different locations within Minnesota over 3 years (Yates and Cochran, 1938). Although homogeneity among the variance components is not assured, the conditions of this example should provide for a more uniform degree of homogeneity than would those for combining independent grazing trials across a broader population.

Combining data from a series of grazing trials tends to lead to a class of models referred to as the nonhomogeneous error models (Giesbrecht, 1989b). Extremely heterogeneous variance components, or discrepant error mean squares, can make the task of pooling datasets more difficult. These heterogeneous components may indicate a lack of full randomization, use of different protocols by independent researchers, or some other departure from the strict intentions of the experimental design. For a particular response across all studies, the magnitude of heterogeneity may be so dramatic that it would be erroneous to analyze the datasets using pooled variance components.

In the previous combined analysis of variance example (series of **k** grazing trials to estimate the effect of a treatment using an experimental /control group design), the estimates of errors from all trials are pooled. If the residual errors of all trials are similar, such pooling provides a more accurate estimate than the estimates derived from the independent trials, because a larger number of error degree of freedom is available. If the errors are different, the pooled estimate of error variance is an estimate of the mean of the error variances of the separate experiments (Yates and Cochran, 1938). Therefore, it will still be the correct estimate of the error affecting the mean difference of the treatments over all trials, but it will no longer be applicable to comparisons involving some of the trials only.

Experimental error variances can be tested to determine if they differ significantly from trial to trial by Bartlett's test for homogeneity of variances (Snedecor and Cochran, 1967). Some factors may give stable responses from trial to trial, while others may be more variable in their performance.

Combined Analyses in Grazing Research

An extensive review of the literature indicated that there have been few attempts to combine either raw data or results from independent but similar grazing trials (Petersen and Lucas, 1960). This was expected, since most of the emphasis has been placed on the design and analysis of new grazing experiments. I believe that the lack of publications describing methods for combining data from grazing trials can be attributed to: 1) the greater appeal and demand for conducting trials that use newly developed forages and systems of animal management, 2) the time-consuming task of concatenating several datasets, 3) the high level of statistical skill or access to consultation needed to analyze appropriately these datasets, and 4) the previous lack of powerful computing resources to perform complex matrix operations with relative ease.

Grazing trials are unwieldy, expensive and the number of treatments that can be studied in a given trial is limited. These facts may have contributed to the lack of combined analyses in grazing research because there does not exist a large collection of independent trials which have addressed the same questions. The recent identification of the immense tall fescue (*Festuca arundinacea* Schreb.) toxicosis problem in the eastern US has led to the initiation of several grazing trials that have examined similar tall fescue treatments using comparable livestock and grazing methodologies.

McIntosh (1983) provided several examples of analysis of variance tables for combined experiments which could be applied directly to grazing trials replicated over locations and/or years. Fisher F-ratios for fixed, random, and mixed models are listed in each example analysis of variance table.

The Petersen and Lucas (1960) paper was one of the first and is one of the most often cited research papers on combining grazing trials. In it, they defined how one can use a number of unrelated experiments to estimate parameters that are not estimable in individual grazing trials. They developed a model for the components that made up the experimental errors of grazing trials. They reported that the most important sources of variability in animal performance are the between-animal variance, the animal X time interaction, and the pasture X time interaction. Important sources of variability for liveweight production ha^{-1} were the pasture X time and the animal X time interactions.

To estimate the magnitude of experimental error on a pasture basis for average daily gain (ADG), they obtained data from 40 grazing seasons from replicated grazing trials at 10 experiment stations in the southwestern and midwestern US. Eight trials were conducted with beef cattle, the remaining two with sheep (*Ovis* L.). The pastures were composed of improved, humid region species.

From these data, the per pasture variances for ADG were estimated by an analysis of variance. Estimates of the between pasture variability in herbage quality as measured by animal performance, pasture X time interaction for herbage quality, between-animal variability in performance, and the animal X time interaction for performance, were estimated using ordinary least squares.

For ADG, it was found that the pasture component contributed a negligible amount to experimental error. They concluded that the magnitude of variation among animals should vary inversely as the number of animals and the time spent by each animal on the pasture.

Similar equations to the one developed by Petersen and Lucas (1960) for the experimental error associated with ADG were not developed for describing the experimental error associated with liveweight production ha^{-1} or animal grazing days ha^{-1}. Examination of the errors computed from the trials indicated that the experimental error of product ha^{-1} was of the same magnitude as, but tended to be lower than, that of ADG.

Although the Petersen and Lucas method is not the most efficient method for combining evidence, according to Giesbrecht (1989a), it is still a valid and useful technique that is easy to understand. On the other hand, the current advances in statistical theory have allowed for more accurate methods of analyses. When raw data are obtainable, the general purpose regression or least squares programs available in most statistical software packages can make a combined analysis feasible. An important classical assumption is that the various datasets have a common variance. One must assume also that there are no other random factors in the model(s) in addition to the residual error term (Giesbrecht, 1989a).

A generalized least squares analysis must be used if one is not willing to assume that the errors are homogeneous and/or if there is a more complex random error structure (Giesbrecht, 1989a). Burns et al. (1983) provide an example of this type of analysis. They evaluated a two-step statistical procedure to analyze cow-calf responses from an unbalanced grazing trial in which treatments were deleted or added as the study progressed. The statistical procedure involved a first step of estimating variance components for year and pasture effects. These were applied in a second step as a weighted adjustment through a generalized least squares analysis. The variance components, treatment mean adjustments and associated standard deviations of the treatment means from the two-step analysis were compared with two ordinary least squares analyses. In the first ordinary least squares analysis, years were defined as fixed effects and the effect of pastures was ignored. In the other ordinary least squares analysis, pastures were defined as fixed effects and the effect of years was ignored. The residual errors from the ordinary least squares analyses were consistently larger than those of the two-step analysis. They concluded that two-step analysis gave biologically rational adjustments of treatment means and offered much potential for experiments where unbalanced data were likely in treatment evaluation.

Burns et al. (1983) further concluded that fair comparison of treatments evaluated in the unbalanced dataset could occur if the means were adjusted for both year and pasture effects. The two-step procedure allowed years and pastures to be interpreted as combining information within and among pastures, and within and among years. Specific variables in question may also have differing variance components. For example, they determined that ADG was influenced less by pastures than was liveweight production ha^{-1}, while both showed a similar year effect.

Henderson's Mixed Models Method

The statistical methods used most often in agricultural research are based on regression, the analysis of variance, or the analysis of covariance (Stroup, 1989). The appropriate techniques for balanced random or fixed effects models are established thoroughly in the statistical literature. There is also a wealth of literature describing mixed model procedures but they are often under-used by most agricultural scientists.

A mixed model is defined as a model in which some of the effects are fixed and some are random (Searle, 1971). In truth, all models are actually mixed models, because they all contain a fixed μ and a random error term; the mixed model description, however, is used commonly for models where effects other than μ and the residual errors are a mixture of fixed and of random effects.

It is well known that most analyses of data from agricultural experiments have some mixed model aspect. Henderson's (1975b) work on 'best linear unbiased prediction' (BLUP) in animal breeding represents the best known and possibly the most successful use of mixed models methods. Henderson's mixed model can be portrayed in matrix form by the following equation:

$$\mathbf{Y} = \mu + \mathbf{X}\beta + \mathbf{ZU} + \epsilon \qquad (1)$$

where:

Y is an n x 1 vector of measured responses (*e.g.*, ADG),

μ is the overall mean of the measured response (*e.g.*, mean ADG),

X is an n x p design matrix of fixed effects (*e.g.*, treatments, sex),

β is a p x 1 vector of unknown fixed effects to be estimated (*e.g.*, estimated effect of endophyte-free (E-) tall fescue, endophyte infected (E+) tall fescue, *etcetera*),

Z is an n x q design matrix of random effects (*e.g.*, blocks, locations, years, and all possible random and random-fixed interactions),

U is a q x 1 vector of unknown random effects, with a mean of zero, to be predicted (*e.g.* predicted effect of a specific location), and

ϵ is an n x 1 error vector with a mean of zero.

This mixed model assumes that the variances and covariance of the vectors U and ϵ are given by the matrices G and R, respectively.

$$\text{Variance}\begin{bmatrix} U \\ \epsilon \end{bmatrix} = \begin{bmatrix} G & 0 \\ 0 & R \end{bmatrix} \qquad (2)$$

In grazing research, the G matrix could be comprised of the location, year, and block relationships among pastures or animals. In the context of most grazing research, the R matrix is equal to the variance/covariance among residual errors. In cases where there is no correlation among the random effects of G and the random errors of R, or the correlation is simply ignored, both matrices are diagonal and are easily invertible. Henderson demonstrated that estimates of β and U (b and u, respectively) can be obtained from any solution of the following equations:

$$\begin{bmatrix} X'R^{-1}X & X'R^{-1}Z \\ X'R^{-1}X & Z'R^{-1}Z + G^{-1} \end{bmatrix} \begin{bmatrix} b \\ u \end{bmatrix} = \begin{bmatrix} X'R^{-1}Y \\ Z'R^{-1}Y \end{bmatrix} \qquad (3)$$

Solving for the unknown vector yields the equations:

$$\begin{bmatrix} b \\ u \end{bmatrix} = \begin{bmatrix} X'R^{-1}X & X'R^{-1}Z \\ X'R^{-1}X & Z'R^{-1}Z + G^{-1} \end{bmatrix}^{-1} \begin{bmatrix} X'R^{-1}Y \\ Z'R^{-1}Y \end{bmatrix} \qquad (4)$$

The solution of Henderson's mixed model equations provides a solution for which the estimate of the b fixed effects is equal to that obtained by generalized least squares, or 'best linear unbiased estimates' (BLUE). In addition to BLUE, Henderson's mixed models formulate the random portion into one of estimation of realized values of random variables, the random variables being the elements of the U vector. This technique is known as 'best linear unbiased prediction' (BLUP).

McLean (1989) provided a comparison between fixed and mixed model methodology where he showed the differences in technique and principles. He also provided some of the important properties of BLUP solution of the mixed model equations.

Sanders (1989) discussed the merit and intent of six different models in combined analyses using 32 soybean [*Glycine max* (L.) Merr.] entries planted in an incomplete block design with 6 entries per block at 2 distinctly different locations (environments). Models were defined to make predictions for individual experiments, individual locations from combined data, or for entries over all locations, depending upon the desired inference space. The variance components were pooled in some of the models; in other models, the heterogeneity among locations, among blocks, among blocks by entries, and residual error was accounted for in the G and R matrices. Generalized least squares means of soybean yields were similar for the analyses which weighted and did not weight the random effects variance components of each location into the **G** matrix and the residual variance components into the **R** matrix.

Stroup (1989) gave a description of the 'shrinkage estimator' of BLUP using batting averages of professional baseball players on three dates in 1985. He showed that BLUP from early season batting averages were better predictors of the entire-season batting averages than BLUE from late-season batting averages. The shrinkage estimator, more commonly referred to as 'regression toward the mean', occurs when the conditional mean of the upper or lower portion of the distribution shifts, or reverts, toward the unconditional mean μ (Samuels, 1991).

Hill and Rosenberger (1985) showed that BLUP was superior to other methods of combining unbalanced data for estimating mean yields of alfalfa (*Medicago sativa* L.) genotypes from a series of evaluations at one location over a period of eight years. Bridges (1989) showed that BLUP was superior to BLUE in predicting the mean yields of four cucumber (*Cucumis sativus* L.) cultivars evaluated at two locations.

Predictable functions and prediction spaces may be one of the more important features of Henderson's mixed models procedure. Stroup (1989) demonstrated the flexibility of predictable functions, or the mixed model generalization of estimable functions. Predictable functions are defined to obtain least-squares means, and differences among treatment means or groups of treatments means, depending on the implied prediction or inference space. Inference space is defined as the set of elements, or population, to which the prediction function is intended to apply (Stroup, 1989).

There are three general predictable functions and, in turn, inference spaces, in Henderson's mixed models. Broad predictable functions do not retain any of the coefficients of the random effects in the estimation of the treatment effects. They are predictors of a fixed effect(s) applied to the entire population represented by the experimental data. The SE of these functions involves variance components of all the random effects. Intermediate predictable functions retain some of the coefficients of the random effects. The SE of these functions include the variance components of those terms which were excluded in the predictable function and the residual error term. Those coefficients which were retained in the intermediate predictable function have the same effect on the SE as regarding them as fixed effects. Narrow predictable functions retain all of the coefficients of the random effects in the estimation of treatment effects. Narrow predictable functions have an inference space similar to the traditional fixed effect estimable functions of ordinary least squares procedures -- the inference space is specific for the spectrum of locations, years, blocks, *etcetera* observed in the experiment.

Conventional linear model computing software packages, *e.g.*, SAS™ GLM procedure, use only the narrow form of the predictable function to compute least squares means, contrasts, *etcetera*. Most default to SE appropriate for the narrow case. Standard errors for the intermediate inference space can be obtained by specifying optional error terms other than the residual error term. No analogy to the broad inference space exists in conventional linear model software (Stroup, 1989). It would seem that most researchers would be more interested in obtaining predictions of livestock performance for future years and different pastures rather than for a more finite population which is restricted to the years and pastures when and where data were collected. The broad and intermediate predictable functions simply provide more meaningful predictions of future performance than do narrow predictable functions.

Therefore, a major advantage of Henderson's mixed models is that one can formulate a prediction or inference space for characteristics in the future, *e.g,* future livestock performance or future forage productivity. A broad inference space estimate could be the mean steer response to tall fescue treatments for all southeastern locations and future grazing years, and for different pastures of similar management and botanical composition. An intermediate inference space estimate could be the mean steer response at a few locations over all grazing years. A narrow inference space estimate could be the mean steer response at one location during the spring of 1979.

One of the limitations of this procedure is the present shortage of computer software to perform the analysis. The General Linear Mixed Models (GLMM) software is currently available, and with it one can perform a mixed model analysis with relative ease (Blouin and Saxton, 1990). On the other hand, this software does not allow for the incorporation of heterogeneous variance components into the G and R matrices. The interactive matrix language (IML) of SAS™ (1985a) has been used to write the instructions that are necessary to weight the individual variance components of locations, years, *etcetera* in the G matrix and the residual errors in the R matrix (Panter, 1991).

CHAPTER 2

RATIONALE AND JUSTIFICATION

Tall Fescue Toxicosis Problem

Detrimental effects on livestock performance due to consumption of tall fescue (*Festuca arundinacea* Schreb.) pastures infested with the endophytic fungus (*Acremonium coenophialum* Morgan-Jones & Gams) have been documented widely since the fungus was first recognized as a causal agent of tall fescue toxicosis (Stuedemann and Hoveland, 1988; Fribourg et al., 1991a). This problem is serious in the transition zone of the southeastern US, affecting over 14 million ha of pasture land. Economic losses attributed to tall fescue toxicosis are over $600 million for beef (*Bos spp.* L.) cattle alone (Hoveland, 1991). Several studies have shown that inclusion of legumes (*Fabaceae* L.) into infested sods or the use of endophyte-free (E-) cultivars have resulted in improved animal performance (Stuedemann and Hoveland, 1988; Fribourg et al., 1991b). It has also been demonstrated that E- tall fescue is less persistent and pest resistant than is infected (E+) tall fescue (Bacon and Siegel, 1988).

The literature does indicate that livestock performance from either E+ or E- pastures, with or without legumes, is somewhat consistent throughout the transition zone. On the other hand, livestock performance obtained at certain locations (environments) is quite variable within the transition zone. The reasons for the variability in livestock performance among locations have not been addressed. Therefore, it may be valuable to determine why livestock performance is inconsistent among groups of environments. The magnitude of the tall fescue toxicosis problem and the lack of precise knowledge concerning the effect of the endophyte on livestock also warrant that research be continued in this area.

Individual Versus Multiple Tall Fescue Grazing Trials

Most of the information concerning the effect of the endophyte on livestock performance was obtained from grazing trials conducted as discrete independent endeavors. Repetition is a common characteristic of many of these independent tall fescue grazing trials. The differences in soils, in some agronomic practices, in climatic situations or seasons of the year, and in other variations in environmental conditions have warranted the repetition of the same treatments throughout the zone of tall fescue adaptation. It is well known that tall fescue pasture and animal management are often site specific. Therefore, an independent trial conducted at one location is justified because it permits inferences to be made for a specific, finite population, *e.g.*, E- tall fescue plus ladino clover pasture management for a group of several counties in the Coastal Plain physiographic region of West Tennessee.

On the other hand, the results obtained at a single location during a single year or for two to three consecutive years, however accurate themselves, are of limited utility, either for the immediately practical end of determining the most profitable forage combinations, stocking rates, *etcetera*, or for the more fundamental task of elucidating the underlying scientific principles. The results obtained from an independent study may or may not be applicable to a larger population, *e.g.*, West Tennessee, depending on whether the experimental site encompassed the diversity of soils found in this region and whether the length of grazing included a wide spectrum of the climatic conditions that occur in this region. I believe that the implied population of inference in a tall fescue grazing trial should be all tall fescue pastures to which application of the treatment is contemplated, and that this population should be as large as possible.

As progress is made in forage-livestock systems, investigators look for smaller effects, typically without the concomitant increases in budgets required for more sensitive grazing trials. The large expense of land, labor, animals, and facilities has caused a general restriction in the number and kinds of objectives. It is possible that the most profitable forage-animal system for a given population was never included in a single grazing trial. The small number of experimental units (pastures and/or animals) and replications, and the relative short durations of grazing, constitute a common, unfortunate characteristic of many grazing trials.

Grazing trials are simply limited by financial and logistical constraints that restrict the duration and number of treatments to be investigated. Consequently, a major weakness of most independent grazing trials is a shortage of error degrees of freedom (df) and the associated lack of power for detecting small differences within a group of treatments. It has been well documented that, when faced with a number of small, independent studies, there is a very strong tendency to conclude that small but real effects are nonexistent (Hedges and Olkin, 1985). The lack of power associated with independent grazing trials may be a major reason why there is a moderate degree of non-uniformity in results obtained from separate trials using similar tall fescue treatments and livestock.

Analysis of Groups of Experiments

One way to understand better the complex forage-livestock interface over a large population, *e.g.*, the transition zone (southern Illinois and Ohio south to northern Mississippi and Georgia, eastern Oklahoma east to the Piedmont in Virginia and the Carolinas) of the eastern US, is to combine data from independent but comparable tall fescue grazing trials. Pooling data from these trials would result in a greater number of error df and increased power for making sound, statistical inferences than conducting separate analyses. Small but real statistical differences that were not detectable in the primary analysis of each of a number of given grazing trials may be revealed in a secondary analysis of concatenated datasets.

Results obtained from the combined analyses could be used to make forage and animal management recommendations for a larger population or set of populations, since the studies would reflect forage and animal responses over diverse locations, soils, years, climatic conditions, and kinds of animals. In some cases, it may be erroneous to make recommendations for the entire population, the transition zone, for reasons already discussed. Animal performance and forage productivity obtained from each tall fescue treatment should probably be ranked for each location.

Combined analyses also may provide the means to quantify the variability in forage productivity and animal performance among tall fescue treatments, soils, and climatic conditions across and within locations and years. One should remember that the primary purpose of an analysis of variance is to produce estimates of one or more error mean squares, and not (as is often believed) to provide significance tests (Finney, 1989a). Combining data from a group of experiments would allow for the comparison of these components of variation. This in itself would be a worthy contribution, since there are limited data available on the sources and magnitudes of the variance components from tall fescue grazing trials specifically, and from grazing trials generally. Knowledge of the magnitude of these variance components would aid researchers in the experimental design of future grazing trials. The degree of power for detecting significant differences on these kinds of forage-livestock systems could be inferred.

Quantifying these components may also be of value to modelers of forage-livestock systems. Systems analysis has become a useful tool for examining the forage-livestock interface. These components of variation could provide modelers with the coefficients needed to simulate data. Combining these studies could be viewed also as a forage-livestock interface model for the transition zone.

Comparability among different studies is often difficult because of the difficulty in achieving complete objectivity in grazing trials. The combined analysis also may provide insight into the reasons for some lack of agreement among the results of independent tall fescue grazing trials. The difficulties are not restricted to grazing trials that often involve personal judgment at various points (Wheeler et al., 1973). One possible view is that this should be accepted as another source of experimental error, error in the level of the treatment (Giesbrecht, 1989a). A combined analysis of livestock performance on tall fescue pastures, where different researchers--each representing slightly different views, methods, and idiosyncrasies--

provide the replication that leads to the measure of experimental error, may in the long run provide us better information. The purpose of research and the publication of the interpretations is to inform others of the possible consequences of future actions.

In conclusion, combining datasets from several independent tall fescue grazing trials would have value to the forage-livestock discipline. Combining data from several grazing trials may help to integrate results so as to uncover patterns of underlying relations and causalities. These in turn will contribute to the establishment of general principles and cumulative knowledge. In the fields of agronomy and animal science, a major need today is for some means to make sense of the vast amounts of data that have been accumulated already, rather than gathering additional empirical data. A combined analysis may help researchers reach more definite conclusions and provide the foundation for further investigations on forage-livestock productivity and management in the transition zone. Scientific advances usually result from the accumulation of knowledge obtained from many studies.

CHAPTER 3

COMBINED ANALYSES OF STEER PERFORMANCE FROM INDEPENDENT TALL FESCUE GRAZING TRIALS

Introduction

Detrimental effects on livestock performance due to consumption of tall fescue (*Festuca arundinacea* Schreb.) pastures infested with the endophytic fungus (*Acremonium coenophialum* Morgan-Jones & Gams) have been documented widely since the fungus was recognized as a causal agent of tall fescue toxicosis (Stuedemann and Hoveland, 1988). This problem is serious in the transition zone (southern Illinois and Ohio south to northern Mississippi and Georgia, eastern Oklahoma east to the Piedmont in Virginia and the Carolinas) of the eastern US, affecting over 15 million ha (Buckner et al., 1979). Economic losses are estimated to be over $600 million for beef (*Bos* spp. L.) cattle alone (Hoveland, 1991). Several trials have shown that inclusion of legumes (*Fabaceae* L.) into infested (E+) sods or the use of endophyte-free (E-) cultivars has resulted in improved animal performance (Stuedemann and Hoveland, 1988; Fribourg et al., 1991a). Endophyte-free tall fescue is less persistent and pest resistant than is E+ tall fescue (Read and Camp, 1986; Bacon and Siegel, 1988).

Most of the information concerning the effects of the endophyte on livestock performance was obtained from grazing trials conducted as discrete endeavors. Differences in soils, in agronomic practices, in animal breeds or management, in climatic situations or seasons of the year, and in other environmental conditions, have warranted the repetition of similar treatments in tall fescue experiments throughout the transition zone. On the other hand, the results obtained at a single location during a single year or for two or three consecutive years, however accurate themselves, are of limited utility, either for the immediately practical end of determining the most profitable forage combinations, stocking rates, *etcetera* or for the more fundamental task of elucidating the underlying scientific principles.

An extensive review of the literature indicated that there have been few attempts to combine either raw data or results from independent but similar grazing trials (Petersen and Lucas, 1960). This was expected, since most of the emphasis has been placed on the design and analysis of new grazing experiments. The small number of publications describing methods for combining data from grazing trials may be attributed to: 1) the greater appeal and demand for conducting trials that use newly developed forages and systems of animal management, 2) the time-consuming task of concatenating several datasets, 3) the high level of statistical skill or access to consultation needed to analyze appropriately these datasets, and 4) the previous lack of computing resources powerful enough to perform complex matrix operations with relative ease.

Grazing trials are unwieldy and expensive, and the number of treatments that can be studied in a given trial is limited. These facts may have contributed to the lack of combined analyses in grazing research, because there does not exist a large collection of independent trials which have addressed the same questions. The recent identification of the extensive tall fescue toxicosis problem in the eastern US has led to the initiation of several trials that have examined similar tall fescue treatments using comparable livestock and grazing methodologies.

As progress is made in forage-livestock systems, investigators look for smaller effects, typically without the concomitant increases in budgets required for more sensitive grazing trials. Grazing trials are usually limited by financial and logistical constraints that restrict the duration and the number of treatments to be investigated (Bransby, 1989). The small number of experimental units (pastures and/or animals) and replications, and the short durations of grazing, constitute a common characteristic of grazing trials. Consequently, a major weakness of most independent grazing trials is a shortage of error degrees of freedom and the associated lack of power for detecting small differences within a group of pasture treatments. It has been well documented that, when faced with a number of small, independent studies, there is a very strong tendency to conclude that small but real effects are nonexistent (Hedges and Olkin, 1985). The lack of power associated with independent grazing trials may be a major reason why there is a moderate degree of non-uniformity in results obtained from separate studies using similar tall fescue treatments and livestock.

Comparability among different grazing trials is often difficult because of the problem of achieving complete objectivity in grazing research. It is possible that this should be accepted as another source of true experimental error, error in the level of the treatment (Giesbrecht, 1989a). A combined analysis of beef (*B. taurus* L.) steer performance on tall fescue pastures, where different investigators -- each with slightly different convictions, methods, and environments -- provide the replication that leads to the measure of experimental error, in the long-run should give the forage-livestock discipline better information.

Henderson's Mixed Models procedure (Henderson, 1975a,b; 1984) provides the opportunity to combine several related grazing trials to estimate performance of steers grazing tall fescue pastures over a broad inference space (McLean et al., 1991) of future grazing years and different tall fescue pastures of similar botanical composition and management. This procedure allows the variance components of the random effects of years, replications, locations, and all random-random and random-fixed interactions to be incorporated into the estimates of the fixed effect of tall fescue treatments (Stroup, 1989).

The objectives of this study were: 1) to provide combined estimates of steer performance on E+ and E- tall fescue pastures in the eastern US, with and without clover, 2) to determine the functional relationships between steer ADG and endophyte infestation level, and 3) to demonstrate the effectiveness and flexibility of Henderson's mixed model procedure in the combined analyses of grazing trials. Results obtained from the combined analyses can be used to characterize steer performance and variability over a diverse set of locations, soils, years, and climatic conditions.

Materials and Methods

The Data Base

Datasets were pooled from 12 tall fescue grazing trials conducted during the last 13 years at nine locations in seven southeastern or adjoining states (Figure 1). This collection was the most inclusive experimental data of steer performance on tall fescue that could be assembled. All dataset contributors provided, from personal communications and published reports, complete summaries of the objectives, experimental methodology, description of soils, animal breeds, durations, experimental design used

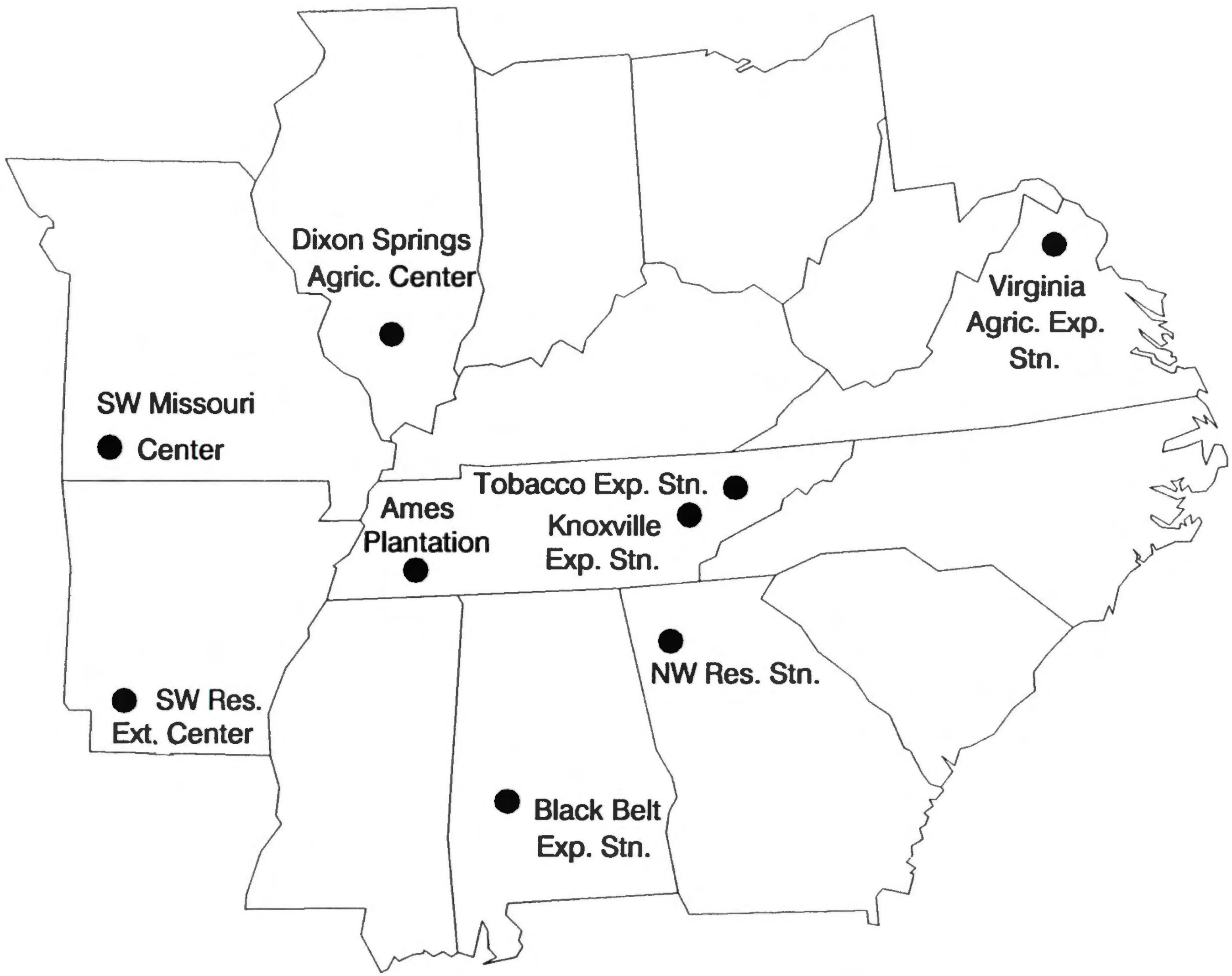

Figure 1. Location of independent grazing trials used in the combined analyses of livestock performance on endophyte infected and not infected tall fescue.

including number and sizes of pastures, *etcetera* for their respective studies [Tables 1 to 6: Hoveland et al., 1983; Mitchell et al., 1986; Pedersen et al., 1986; Schmidt et al., 1986; Phillips et al., 1990; Chestnut et al., 1991; Fribourg et al., 1991b; Thompson et al., 1991,1992; Allen et al., 1992]. A summary of the available treatments and the results of each grazing year within locations was prepared. Forage treatments other than tall fescue, with and without clover, were deleted from the data base. Although the Middleburg, VA and the Mount Vernon, MO datasets included both steers and heifers, the data for heifers were used for computing pasture carrying capacity but not for daily gains.

Tall Fescue Treatments

Forage treatments included tall fescue at three ranges of endophyte infestation, either with or without clover:

EF	Endophyte-free, ≤ 5% E+
ME	Moderately infested, ≥ 20% to ≤ 35% E+
HE	Highly infested, ≥ 50% to ≤ 97% E+
EFCL	Endophyte-free, ≤ 5% E+, plus clover
MECL	Moderately infested, ≥ 20% to ≤ 35%, plus clover
HECL	Highly infested, ≥ 50% E+ to ≤ 97%, plus clover

Table 1. Tall fescue treatments available for analysis in each spring grazing trial in seven states.

Locations	State	Years	Tall Fescue Treatments Without clover[+]			With clover[§]		
			EF	ME	HE	EFCL	MECL	HECL
Black Belt Exp. Stn., Marion Junction	AL	1979-83	EF		HE			
Black Belt Exp. Stn., Marion Junction	AL	1981-83		ME				
SW Res. and Ext. Center, Hope,	AR	1986-87	EF		HE	EFCL		HECL
NW Research Station, Calhoun	GA	1985; 1987-88	EF			EFCL		
Dixon Springs Agric. Center, Simpson	IL	1989; 1991			HE			HECL
SW Missouri Center, Mount Vernon	MO	1988-90	EF		HE			
Ames Plantation, Grand Junction	TN	1979-81					MECL	
Ames Plantation, Grand Junction	TN	1983-85	EF		HE	EFCL		HECL
Ames Plantation, Grand Junction	TN	1987-89				EFCL	MECL	HECL
Knoxville Experiment Station	TN	1990-91	EF	ME	HE			
Tobacco Experiment Stn., Greeneville	TN	1985-89					MECL	HECL
Virginia Agric. Exp. Stn., Middleburg	VA	1983-88			HE			HECL

[+] EF = endophyte free; ME = moderately infested; HE = highly infested.

[§] EFCL = endophyte free + clover; MECL = moderately infested + clover; HECL = highly infested + clover.

Table 2. Tall fescue treatments available for analysis in each summer and spring+summer grazing trial in two states.

Locations	State	Years	Tall Fescue Treatments Without clover[+]			With clover[§]		
			EF	ME	HE	EFCL	MECL	HECL
NW Research Station, Calhoun	GA	1985	EF			EFCL		
Ames Plantation, Grand Junction	TN	1979-81					MECL	
Ames Plantation, Grand Junction	TN	1983-85	EF		HE	EFCL		HECL
Ames Plantation, Grand Junction	TN	1987-89				EFCL	MECL	HECL
Knoxville Experiment Station	TN	1990-91	EF	ME	HE			
Tobacco Experiment Stn., Greeneville	TN	1985-89					MECL	HECL

[+] EF = endophyte free; ME = moderately infested; HE = highly infested.

[§] EFCL = endophyte free + clover; MECL = moderately infested + clover; HECL = highly infested + clover.

Table 3. Tall fescue treatments available for analysis in each fall+winter grazing trial in three states.

Locations	State	Years	Tall Fescue Treatments Without clover[+]			With clover[§]		
			EF	ME	HE	EFCL	MECL	HECL
Black Belt Exp. Stn., Marion Junction	AL	1978-83	EF		HE			
Black Belt Exp. Stn., Marion Junction	AL	1980-83		ME				
Ames Plantation, Grand Junction	TN	1987-90				EFCL	MECL	HECL
Knoxville Experiment Station	TN	1990	EF	ME	HE			
Virginia Agric. Exp. Stn., Middleburg	VA	1982-87			HE			HECL

[+] EF = endophyte free; ME = moderately infested; HE = highly infested.

[§] EFCL = endophyte free + clover; MECL = moderately infested + clover; HECL = highly infested + clover.

Table 4. Summary of the soil series and families for each of the locations used in the combined analyses.

Locations	Series	US Soil Family
Black Belt Exp. Stn., Marion Junction, AL	Eutaw	very fine, montmorillonitic, thermic Entic Pelluderts
	Sumter	fine-silty, carbonatic, thermic Rendollic Eutrochrepts
	Houston	very fine, montmorillonitic, thermic Typic Chromuderts
SW Res. and Ext. Center, Hope, AR	Sawyer	fine-silty, siliceous, thermic Aquic Paleudults
NW Research Station, Calhoun, GA	Townley	clayey, mixed, thermic Typic Hapludults
Dixon Springs Agric. Center, Simpson, IL	Grantsburg	fine-silty, mixed, mesic Typic Fragiudalfs
	Zanesville	fine-silty, mixed, mesic Typic Fragiudalfs
SW Missouri Center, Mount Vernon, MO	Keeno	loamy-skeletal, siliceous, mesic Mollic Fragiudalfs
	Hoberg	fine-loamy, siliceous, mesic Mollic Fragiudalfs
	Gerald	fine, mixed, mesic Umbric Fragiaqualfs
Ames Plantation, Grand Junction, TN	Memphis	fine-silty, mixed, thermic Typic Hapludalfs
Knoxville Experiment Station, TN	Decatur	clayey, kaolinitic, thermic Rhodic Paleudults
	Dewey	clayey, kaolinitic, thermic Typic Paleudults
Tobacco Experiment Stn., Greeneville, TN	Dummore	clayey, kaolinitic, mesic Typic Paleudults
	Dewey	clayey, kaolinitic, thermic Typic Paleudults
	Etowah	fine-loamy, siliceous, thermic Typic Paleudults
Virginia Agric. Exp. Stn., Middleburg, VA	Chester	fine-loamy, mixed, mesic Typic Hapludults
	Eubanks	fine-loamy, mixed, mesic Typic Hapludults
	Bowmansville	fine-loamy, mixed, nonacid, mesic Aeric Fluvaqents
	Warsham	clayey, mixed, thermic Typic Orchraquults
	Brandywine	sandy skeletal, mixed, mesic Typic Dystocrept

Table 5. Summary of the sexes, breeds, and initial weights of the animals for each location used in the combined analyses.

Locations	Sex[+]	Breeds	Initial Weight
			- kg -
Black Belt Exp. Stn., Marion Junction, AL	S	Angus, Hereford, Shorthorn Crossbreds	244
SW Res. and Ext. Center, Hope, AR	S	Hereford-Brahman X Simmental; Brangus X Simmental	285
NW Research Station, Calhoun, GA	S	Angus, Hereford, Shorthorn Crossbreds	219
Dixon Springs Agric. Center, Simpson, IL	S	Hereford; Angus; Hereford X Angus	236
SW Missouri Center, Mount Vernon, MO	S & H	Simmental, Hereford, Angus, Charolais, and Limousin Crossbreds	243
Ames Plantation, Grand Junction, TN	S	Angus	236
Knoxville Experiment Station, TN	S	Angus; Angus Crossbreds	264
Tobacco Experiment Stn., Greeneville, TN	S	Angus; Hereford X Angus	244
Virginia Agric. Exp. Stn., Middleburg, VA	S & H	Angus	252

[+] S = steers; H = heifers.

Table 6. Summary of pasture sizes (experimental unit), *Acremonium coenophialum* infestation levels, and tall fescue cultivars for each location used in the combined analyses.

Locations	Pasture sizes (ha)	Infestation[+] level (%)	Cultivars Infested	Cultivars Endophyte-free
Black Belt Exp. Stn., Marion Junction, AL	1.62	≤ 5, 34, and 90	KY 31	AU-Triumph, KY 31
SW Res. and Ext. Center, Hope, AR	1.62	≤ 5, and 80	KY 31	KY 31
NW Research Station, Calhoun, GA	1.58-2.19	≤ 5, and 90	KY 31	AU-Triumph
Dixon Springs Agric. Center, Simpson, IL	2.43-3.48	70	KY 31	
SW Missouri Center, Mount Vernon, MO	0.41	≤ 5, and 60-70	KY 31	Martin, Mozark, Phyter, MO96, Johnstone, KY 31 LLAE[§] and HLAE[§]
Ames Plantation, Grand Junction, TN	1.20	≤ 5, 22, and 35-81	KY 31	AU-Triumph, KY 31 Kenhy, and Johnstone
Knoxville Experiment Station, TN	1.20	≤ 5, 30, and 90	KY 31	AU-Triumph, KY 31 Johnstone, and Martin
Tobacco Experiment Stn., Greeneville, TN	1.20	70 35	KY 31 Forager[†]	
Virginia Agric. Exp. Stn., Middleburg, VA	0.80-1.60	28-55	KY 31	

[+] Pastures ≤ 5%, > 5% and ≤ 35 %, or > 50% infested were coded as endophyte free, moderately infested, or highly infested tall fescue treatments, respectively.

[§] LLAE and HLAE are low and high leaf area experimental varieties (David Sleper, personal communication).

[†] Mixture of endophyte free Forager and highly infested KY 31 remaining from a previous study.

These ranges of endophyte infestation were established based on the findings of Fribourg et al. (1991b). These researchers determined that there was little difference in animal performance when steers grazed tall fescue with 35% or more E+, but the effect of 22% E+ was intermediate between those of 3 and of 35% E+.

Livestock performance obtained from most E- cultivars has been similar; most of the differences in E- cultivars have been due to variabilities in stand persistence and available herbage mass (Fribourg et al., 1991b). There were only a few trials which included different E- cultivars (Table 6). Consequently, similarly performing E- cultivars among and within locations were pooled. 'Kentucky 31' was the only E+ cultivar, except at Greeneville, TN where the ME pastures included a 35% mixture of predominantly E- 'Forager' and some highly infested Kentucky 31 remaining from a previous study.

The tall fescue plus clover pastures contained about 20 to 40% clover in the spring and fall+winter, and about 10% in summer. In the study conducted at Hope, AR, and the 1979-83 and 1983-85 Grand Junction, TN studies, there was about 10 to 35% bermudagrass (*Cynodon dactylon* L.) in the tall fescue pastures during late spring and summer. For the 1979-83 Marion Junction, AL study, there was about 5% bermudagrass and 5% dallisgrass (*Paspalum dilatatum* Poir.) in the tall fescue pastures during late spring and summer. The moderate contamination of tall fescue swards with warm season grasses is typical of these locations.

All treatments were replicated twice in each study. Treatments were arranged in randomized complete block designs (RCB) in each of the five studies conducted in Tennessee. In the studies from the other six states, the treatments were arranged in completely randomized (CRD) designs.

Herbage mass availabilities were not known for all locations. Nevertheless, all dataset contributors indicated that there was at least 1000 kg DM ha^{-1} in the tall fescue pastures available at all times during spring, summer, and fall+winter, implying that steer intake was not limited by availability of forage. Pastures were managed with either put-and-take or set stocking, at rates sufficient to utilize most of the available forage mass without limiting steer intake. A continuous grazing management system was used at all locations. At no time did the steers receive supplemental feed while on pasture, but they all had free access to salt, minerals and water.

Statistical Analyses

An objective evaluation of the effect of E+ and E- tall fescue pastures, with and without clover, on steer ADG was made using the mixed model procedure described by McLean et al. (1991), as implemented in the

General Linear Mixed Models procedure (GLMM) (Blouin and Saxton, 1990). The mathematical model was:

ADG = Location + Year(Location) + Treatment + [Treatment X Location] + [Treatment X Year(Location)] + [Block(Location X Year)] + Initial Weight + Initial Weight^2 + [Animal Grazing days ha^{-1}] + ϵ.

The effect of years was nested within the effect of locations, because the climatic situations at one location were judged not to be representative of those of other locations, *e.g.*, the 1986 climatic situations at Hope, AR were not typical of the 1986 climatic situations at Middleburg, VA. For the RCB-designed Tennessee studies, blocks were uniquely identified. For the other studies arranged as a CRD, the block effect was coded as a value of one for all locations.

In order to estimate steer performance for the treatment X location combinations (intermediate inference space), all model effects were defined as random except the main effects of locations and of treatments, and the interaction of locations and treatments. To estimate mean steer ADG for each treatment across all locations (broad inference space), all effects were defined as random except the main effect of treatments.

The initial weight (IW) of the test steers, the initial weight^2, and the steer grazing days ha^{-1} (GD), were used as covariates. Adjustment of treatment means for IW was done to overcome the effects of balancing steer weights across treatments (Snedecor and Cochran, 1967). For mixed models which included the GD covariate, treatment means were adjusted to estimate steer ADG for pasture treatments at the same stocking rate and the same grazing duration. For mixed models without the GD covariate, steer ADG estimates were partially dependent upon the stocking rate and duration of grazing of a pasture treatment.

Since the covariate initial weight^2 was not significant ($\alpha > 0.15$) in each preliminary combined analysis, it was deleted from the final analytical models used in each combined analysis. Some of the studies were initiated in the fall and continued through the spring (Table 1). For these studies, the spring initial weight was considered to be the steer weight obtained during the first half of March. This was done because steer weights at the initiation of spring grazing were confounded with forage treatments in the studies that lasted from fall throughout the spring. Since each summer study was an extension of its respective spring study, the initial weights were considered as the first weight during spring grazing.

Maximum likelihood (ML) variance components of the random effects were obtained using the VARCOMP procedure of SAS® (SAS, 1985b). Since the VARCOMP procedure does not allow for continuous effects in the model, the linear effects of initial weight and GD were removed using the REG procedure of SAS® -- regression models were analyzed with and without the GD covariate. The ADG residuals were then obtained and analyzed using the VARCOMP procedure. Random effects having ML components set to zero were deleted from the analytical models. The remaining, non-zero variance components were inserted into the GLMM mixed models programs. The GLMM personal computer software does allow for estimation of the random effects components, but the processing time can be increased dramatically if these components are to be solved iteratively.

Estimated differences for specific linear contrasts were predicted along with their associated SE of a difference. To determine the functional relationship between steer ADG and E+ infestation levels, the linear and quadratic effects of E+ incidence were computed for the tall fescue treatments, with or without clover, using nonequally spaced orthogonal polynomials (Gomez and Gomez, 1976). The nonequally spaced linear and quadratic coefficients were obtained using the ORPOL function of the interactive matrix language (IML) of SAS® (1985a). The mean levels of E+ infestation for the endophyte-free, moderate, and the highly infested tall fescue treatments, which were used to obtain the linear and quadratic coefficients, were 2.5, 27.5, and 74%, respectively. A simple linear regression model was also fitted for each season using the GLS treatment means, with or with clover, to provide an estimate of the rate of change in steer ADG as endophyte infestation level increased.

All data were analyzed on a per pasture basis, because the pasture is the experimental unit in most grazing research. According to Snedecor and Cochran (1967), the experimental unit is the smallest, independent subdivision of experimental material. Since steers grazing the same pasture are not independent (*e.g.*, forage consumed by one steer cannot be consumed by another steer), steers were not the appropriate experimental unit. If forage availability or selection are important factors in the response variable, animals within a pasture cannot be considered as independent (Brown and Waller, 1985). Forage availability and selection are probably very important factors when steers are subjected to grazing tall fescue infected with *A. coenophialum*.

Response variables included steer ADG during spring (n = 325), summer (n = 136), spring+summer (n = 136), and fall+winter (n = 124) grazing seasons. Spring grazing was considered to extend from about March 1 to June 30; summer grazing occurred usually from about July 1 to September 1; fall+winter grazing took place from about October 15 to January 30. Although the lengths of seasonal grazing were similar among locations, they were not adjusted to result in identical grazing periods across all grazing years within locations, because the discrepancies in seasonal durations were recognized as a function of the location (environmental conditions) and the grazing year (climatic situations).

Combining datasets from a series of grazing trials tends to lead to a class of models referred to as nonhomogeneous error models (Giesbrecht, 1989b). To determine if the residual errors among locations were homogeneous, the random residual variances were obtained separately for each year within each location, and these were tested using Bartlett's test of homogeneity (Snedecor and Cochran, 1967) for each grazing season. The separate tests of homogeneity indicated that the residual error variances for each grazing year within locations were homogenous among grazing seasons ($\alpha < 0.05$), allowing the error mean squares to be pooled in the combined analyses. The effect of blocks was small ($\alpha > 0.10$) in three of the five Tennessee studies. Therefore, weighting the random effect of blocks(year X location) would probably not have been advantageous.

Results and Discussion

Generalized Least Squares Means of ADG for the Treatments

Implied Inference Spaces for the Treatments

In order to estimate mean ADG for each treatment across all locations, all model effects were defined as random, except the main effect of treatments. Therefore, mean ADG estimates and associated SE for these mixed model analyses represent the broad inference space (McLean et al., 1991) of different tall fescue pastures and locations during future years within the zone of tall fescue adaptation. For the combined spring analyses, the series of locations (environments) and years (climates) represent a random sample of the transition zone, which constitutes the inference space for conclusions concerning treatment effects (Table 1). Since there were fewer datasets for the summer, spring + summer, and fall + winter analyses than there were for the spring analyses, the inference spaces for these seasons are specific for those locations for which data were available (Tables 2 and 3) -- a transitional zone inference space could not be justified considering the geographic representation of summer, spring + summer, and fall + winter seasonal data.

Magnitude of the Standard Errors for the Combined Analyses

The associated standard errors (SE) of the generalized least squares (GLS) treatment ADG means are larger than those of purely fixed models because steer ADG was estimated for the broad inference space of future grazing years and different pastures. The ADG mean estimates and SE of the mean estimates form a frequency distribution for which the means in the combined data base are a random sample of the transition zone.

Steer ADG mean estimates and SE were naturally more variable for those treatments and treatment X location combinations which had fewer pastures (experimental units), replications, and grazing years than did others. The different number of observations affected GLS mean ADG estimates in a statistical sense, but this effect should not be ignored in the interpretation of the combined mixed model analyses -- GLS mean estimates for treatments and locations with fewer observations are considered less reliable. Therefore these estimates tend to revert towards the overall sample mean, and have larger associated SE than do those treatments or treatment X location combinations which had more observations. In these combined analyses, the shrinkage of mean ADG estimates for treatments or treatment X location combinations with fewer observations was a desired property.

Spring Daily Gains

Mean ADG estimates indicate conclusively that steers grazing highly infested tall fescue, with (**HECL**) and without (**HE**) clover, have much smaller daily gains than do those steers grazing endophyte-free tall fescue, with (**EFCL**) and without (**EF**) clover (Table 7). Steers grazing EFCL tall fescue gained 342 and 379 g d^{-1} more than did those steers grazing HECL tall fescue when estimated from models with and without the GD covariate, respectively. Steers grazing EF tall fescue gained 208 and 229 g d^{-1} more than did those steers grazing HE tall fescue when evaluated from models with and without the GD covariate, respectively. This could be due at least partially to the 20% smaller dry matter intake by steers grazing HE tall fescue than by steers grazing EF tall fescue (Chestnut et al., 1991). The effect of moderately infested tall fescue plus clover (**MECL**) was intermediate between those of EFCL and HECL tall fescue-clover mixtures (Table 8).

These results substantiate the consistent findings of the discrete trials. Combining information from similar trials to detect small but real differences was not necessarily imperative for these tall fescue treatments, because it is obvious that the daily gains of steers grazing endophyte-free tall fescue are greater than the daily gains of steers grazing highly infested tall fescue.

Table 7. Spring ADG generalized least squares means for the treatments, estimated from models with and without the steer grazing days ha^{-1} (GD) covariate.

Tall Fescue Treatment	With GD	SE+	No GD	SE+	n§
	---------------- g steer d ----------------				
Endophyte-free	841	55	830	57	98
Moderately infested	757	76	764	81	13
Highly infested	633	54	601	56	66
Endophyte-free + clover	972	63	1022	65	50
Moderately infested + clover	822	67	850	70	32
Highly infested + clover	629	54	644	57	66

+ Broad inference space of different tall fescue pastures of similar endophyte infestation and future years within the transition zone.

§ On a pasture basis with 3 steers or more per pasture.

Table 8. Linear contrasts for steer ADG during the spring grazing seasons, analyzed with and without the steer grazing days ha^{-1} (GD) covariate.

Spring Contrast[+]	Covariate inclusion	Estimated difference	SE[§]	α
		- g $steer^{-1}$ d^{-1} -		
EF vs. ME	With GD	84	64	0.1900
	Without GD	65	68	0.3395
ME vs. HE	With GD	124	66	0.0519
	Without GD	163	70	0.0211
EFCL vs. MECL	With GD	148	68	0.0305
	Without GD	172	73	0.0185
MECL vs. HECL	With GD	194	56	0.0007
	Without GD	206	60	0.0007
EFCL vs. EF	With GD	130	53	0.0154
	Without GD	193	56	0.0006

[+] EF = endophyte-free, ME = moderately infested, and HE = highly infested; EFCL = endophyte-free + clover, MECL = moderately infested + clover, and HECL = highly infested + clover.

[§] Broad inference space of different tall fescue pastures of similar endophyte infestation and future years within the transition zone.

Nevertheless, combining datasets from 12 grazing trials at nine locations within the transition zone provides more meaningful estimates of steer ADG than do those of each independent grazing trial, because there was more information in terms of pastures, years, and locations. The results from the combined analyses also provide a more coherent body of information than do the results obtained from each discrete grazing trial because treatment means were adjusted for the IW and GD covariates.

The combined analyses did detect differences among the six tall fescue pasture treatments which were not obtainable in each separate grazing trial. None of the independent trials included all six of the tall fescue treatments because of financial and logistical constraints and/or the desire to evaluate a group of different endophyte-free cultivars.

Mean ADG estimates were more than 800 g d^{-1} for steers grazing both EFCL and EF tall fescues (Table 7). On the other hand, steers grazing EFCL gained 130 and 193 g d^{-1} more than did those steers grazing EF when estimated from models with and without the GD covariate, respectively (Table 8). The presence of clover in endophyte-free stands may improve steer ADG by providing additional N for increased spring forage and alternative carbohydrates and proteins for the ruminant. Therefore, the inclusion of clover into already productive endophyte-free pastures may be beneficial to producers.

Mean daily gain estimates were similar for steers ingesting EF and ME tall fescue pastures (Table 8). The similarities among ADG estimates for steers grazing these two different kinds of tall fescue was not anticipated. Steers may graze endophyte-free plants preferentially in ME tall fescue stands when forage is not limited, causing spring daily gains to be similar. It is also possible that tall fescue toxicosis does not develop fully in steers ingesting moderately infested ($\geq$ 20% to $\leq$ 35% E+) tall fescue during the cooler conditions of spring.

It has been shown on several occasions that the presence of at least 10 to 25% clover in an E+ stand can alleviate in part the signs of tall fescue toxicosis in animals (Hoveland et al., 1981). On the other hand, spring daily gains of steers grazing HECL and HE tall fescues were virtually identical (Table 7). This suggests that including clover into highly infested tall fescue stands does not ameliorate spring steer performance. Chestnut et al. (1991) suggested that if the relationship between intake of toxic compounds and depressed animal performance is curvilinear (*i.e.*, depression in animal performance with increasing endophyte is greater at low than at high levels of endophyte infestation), then addition of clover into highly infested stands may not dilute intake of E+ tall fescue sufficiently to reduce tall fescue toxicosis. The substitution ratio of clover dry matter for tall fescue dry matter in steer diets may be greater than 1:1 (Goetsch et al., 1987), and therefore clover may not be able to replace sufficient quantities of highly infested tall fescue to reduce tall fescue toxicosis (Chestnut et al., 1991).

Effect of the Initial Weight and Steer Grazing Days Per Hectare Covariates on Spring ADG Estimates

The initial weight (IW) covariate did not explain a substantial portion of the residual variation in spring ADG when analyzed with (α = 0.1042) or without (α = 0.5442) the GD covariate. The IW covariate was almost significant at the 0.10 α level for the mixed model which also included the GD covariate. The partial correlation coefficient between these two covariates, -0.30114, was highly significant (α = 0.0001). This correlation is biologically rational since tall fescue pastures stocked with heavier steers usually have fewer GD.

The GD covariate explained a significant portion of the residual variation in spring ADG (α = 0.0001). Mean ADG estimates were adjusted upwards for pasture treatments that had a larger number of GD than the sample GD mean; the reverse occurred when the mean treatment GD was smaller than the sample GD mean (Table 9).

The differences between ADG means estimated from models with and without the GD covariate were usually small (Table 7). On the other hand, maximum likelihood (ML) variance components for the model with the

Table 9. Initial weights and steer grazing days ha^{-1} (GD) arithmetic means and standard errors (SE) for the tall fescue treatments during spring.

Tall Fescue Treatment	Initial Weight	SE	GD	SE
	-- kg --		-- d ha^{-1} --	
Endophyte-free	260.3	3.65	418.5	20.5
Moderately infested	286.3	3.78	364.4	39.1
Highly infested	265.1	4.85	419.8	28.2
Endophyte-free + clover	237.1	3.67	475.1	23.3
Moderately infested + clover	242.9	3.98	368.9	27.4
Highly infested + clover	250.7	3.91	389.2	26.7

the GD covariate were smaller than those of the model without the GD covariate -- an exception was the variance components for the Block(Year X Location) effect which were essentially equivalent (Table 10).

The size of the random ML variance components provides a guide to the direction and extent of reasonable adjustment of any treatment mean. As expected, the ML variance components for the random effect of years within locations, *i.e.*, the environmental and climatic variabilities, were much larger than the ML variance components of the other random model effects.

Summer Daily Gains

There were fewer treatment differences in summer (Table 11) -- none of the five pre-planned contrasts was significant at the 0.05 α level (Table 12). The generally low daily gains of steers in summer render the detection of differences due to E+ infestation and inclusion of clover more difficult. Nevertheless, there were similarities between summer and spring treatment ADG mean estimates.

As in spring, the mean ADG estimate for steers grazing EF tall fescue was about 180 g d^{-1} larger than the mean ADG estimate for steers grazing HE tall fescue, when estimated from models with and without the GD covariate (Table 11). Steers grazing EFCL gained about 92 g d^{-1} more than

Table 10. Magnitude of the maximum likelihood (ML) variance components for mixed models, with and without the steer grazing days ha^{-1} (GD) covariate, used to estimate mean ADG for the treatments during spring.

Random effect[+]	ML Variance Components[§] With GD	No GD
Year (Location)	59215	63859
Treatment X Location	1797	1992
Treatment X Year (Location)	1397	2057
Block (Year X Location)	12477	12414
Residual Error	18824	20710

[+] The full mathematical model was: ADG = Location + Treatment + [Treatment X Location] + Year (Location) + [Treatment X Year(Location)] + [Block(Location X Year)] + Initial Weight + with or without steer grazing days ha^{-1} + ϵ. All effects were random except the main effect of treatments.

[§] Seasonal ADG variance components not listed from the full model were zero for mixed models with and without steer grazing days ha^{-1}.

Table 11. Summer ADG generalized least squares means for the treatments, estimated from models with and without the steer grazing days ha^{-1} (GD) covariate.

Tall Fescue Treatment	With GD	SE[+]	No GD	SE[+]	n[§]
	---------------- g steer d ----------------				
Endophyte-free	542	78	549	116	30
Moderately infested	525	104	518	139	4
Highly infested	374	86	357	139	10
Endophyte-free + clover	603	78	626	115	32
Moderately infested + clover	578	80	587	116	26
Highly infested + clover	512	77	532	114	34

[+] Broad inference space of different tall fescue pastures of similar endophyte infestation within Tennessee and northwestern Georgia.

[§] On a pasture basis with 3 steers or more per pasture.

Table 12. Linear contrasts for steer ADG during the summer grazing seasons, analyzed with or without the steer grazing days ha^{-1} (GD) covariate.

Summer Contrast[+]	Covariate inclusion	Estimated difference	SE[§]	α
		- g steer^{-1} d^{-1} -		
EF vs. ME	With GD	17	74	0.8165
	Without GD	31	85	0.7168
ME vs. HE	With GD	150	82	0.0721
	Without GD	160	93	0.0877
EFCL vs. MECL	With GD	25	46	0.5920
	Without GD	39	57	0.4995
MECL vs. HECL	With GD	67	37	0.0776
	Without GD	55	46	0.2266
EFCL vs. EF	With GD	61	44	0.1701
	Without GD	77	53	0.1467

[+] EF = endophyte-free, ME = moderately infested, and HE = highly infested; EFCL = endophyte-free + clover, MECL = moderately infested + clover, and HECL = highly infested + clover.

[§] Broad inference space of different tall fescue pastures of similar endophyte infestation and future years within Tennessee and northwestern Georgia.

did those steers grazing HECL. The mean ADG estimate for steers grazing ME tall fescue was virtually identical to the mean ADG estimate for steers grazing EF tall fescue. Steers grazing ME tall fescue gained 150 and 160 g d^{-1} more than did those steers grazing HE tall fescue, when estimated from models with and without the GD covariate, respectively.

In contrast to the spring results, steers grazing EFCL and EF pastures had similar mean daily gains estimates in summer (Table 12). This similarity was expected since the percentage of clover in tall fescue stands declines usually to $< 10\%$ in summer due to inadequate soil moisture, resulting in slower clover growth and possible preferential grazing of clover.

Steers grazing EFCL and MECL pastures had similar summer daily gains estimates (Table 12) -- this effect did not occur in spring or when spring and summer were considered together. Summer ADG estimates of steers grazing the HECL pastures were 138 (with GD) and 175 (without GD) g d^{-1} more than the ADG estimates of steers grazing HE pastures. This was the only seasonal indication that steer ADG is improved by including clover into highly infested ($\geq 50\%$ to $\leq 97\%$ E+) tall fescue stands.

Effect of the Initial Weight and Steer Grazing Days Per Hectare Covariates on Summer ADG Estimates

The IW covariate did not explain a substantial portion of the residual variation in ADG when models were analyzed with (α = 0.0796) and without (α = 0.1104) the GD covariate. The GD covariate explained a significant portion of the residual variation in ADG (α = 0.0029). The arithmetic GD means of the EF, ME, and HE pastures were 325, 334, and 429 d ha^{-1}, respectively. The arithmetic GD means of the EFCL, MECL, and HECL pastures were 269, 244, and 258 d ha^{-1}, respectively.

Hill and Rosenberger (1985) have stated that the mixed model analysis which provides the smallest prediction errors should be preferred. In this study, the SE for the combined analysis with the GD covariate were much smaller than were the SE for the combined analysis without the GD covariate (Table 11).

Daily Gains when considering Spring and Summer Together

Since each summer study was simply an extension of its respective spring study, mixed model analyses were also made to estimate steer ADG for the combined spring plus summer grazing seasons. The spring+summer inference space encompassed steer performance from different tall fescue pastures of similar endophyte infestation, and future grazing years within Tennessee and northwestern Georgia.

Mean daily gains estimates were smaller in spring+summer than they were in spring alone, but the treatment differences were maintained at about the same magnitude (Tables 13 and 14). Steers grazing EF tall fescue gained 183 and 206 g d^{-1} more than did those steers grazing HE tall fescue, when estimated from mixed models with and without the GD covariate, respectively. Steers grazing EFCL tall fescue gained 259 and 275 g d^{-1} more than did those steers grazing HECL tall fescue when evaluated with and without the GD covariate, respectively. The effect of MECL was intermediate between those of EFCL and HECL (Table 14).

The mean spring+summer ADG estimate of steers grazing EFCL pastures was significantly greater than the mean ADG estimate of steers grazing EF pastures (Table 14). Daily gain estimates were similar for steers ingesting EF and ME pastures.

The spring+summer analyses consisted of datasets from only 4 locations within Tennessee and northwestern Georgia whereas the spring analyses contained datasets from 9 locations within 7 eastern states. Therefore, the similarity in treatment effects was not expected, because the spring datasets represented steer daily gains from a much larger population than the spring+summer datasets.

Table 13. Spring+summer ADG generalized least squares means for the treatments, estimated from models with and without the steer grazing days ha^{-1} (GD) covariate.

Tall Fescue Treatment	With GD	SE[+]	No GD	SE[+]	n[§]
	---------------- g steer d		----------------		
Endophyte-free	681	36	689	57	30
Moderately infested	672	63	675	79	4
Highly infested	498	45	483	62	10
Endophyte-free + clover	815	33	855	55	32
Moderately infested + clover	703	33	727	55	26
Highly infested + clover	556	31	580	54	34

[+] Broad inference space of different tall fescue pastures of similar endophyte infestation within Tennessee and northwestern Georgia.

[§] On a pasture basis with 3 steers or more per pasture.

Table 14. Linear contrasts for steer ADG during the spring + summer grazing seasons, analyzed with or without the steer grazing days ha^{-1} (GD) covariate.

Spring + summer Contrast[+]	Covariate inclusion	Estimated difference	SE[§]	α
		- g $steer^{-1}$ d^{-1} -		
EF vs. ME	With GD	9	55	0.8773
	Without GD	14	60	0.8128
ME vs. HE	With GD	174	62	0.0061
	Without GD	192	66	0.0045
EFCL vs. MECL	With GD	112	34	0.0015
	Without GD	128	41	0.0023
MECL vs. HECL	With GD	147	28	0.0001
	Without GD	147	33	0.0001
EFCL vs. EF	With GD	134	34	0.0002
	Without GD	166	37	0.0001

[+] EF = endophyte-free, ME = moderately infested, and HE = highly infested; EFCL = endophyte-free + clover, MECL = moderately infested + clover, and HECL = highly infested + clover.

[§] Broad inference space of different tall fescue pastures of similar endophyte infestation and future years within Tennessee and northwestern Georgia.

Effect of the Initial Weight and Steer Grazing Days Per Hectare Covariates on Spring + Summer ADG Estimates

The IW covariate did not explain ($\alpha > 0.34$) a substantial portion of the residual variation in spring + summer ADG. The GD covariate removed a significant portion of the residual variation in spring + summer ADG ($\alpha = 0.0005$). Daily gains were adjusted upwards for pasture treatments that had a larger number of GD than the sample GD mean. The arithmetic GD means of the EF, ME, and HE tall fescue pastures were 896, 728, and 1127 d ha^{-1}, respectively. The arithmetic GD means of the EFCL, MECL, and HECL tall fescue pastures were 818, 635, and 735 d ha^{-1}, respectively.

Fall Plus Winter Daily Gains

The fall + winter combined analyses included datasets from: 1) Marion Junction, Alabama; 2) Grand Junction, Tennessee; 3) Knoxville, Tennessee; and 4) Middleburg, Virginia. Therefore, the fall + winter inference space represented steer daily gains during future years and from different pastures of similar endophyte infestation among these locations.

Steers grazing EF tall fescue pastures gained 191 and 211 g d^{-1} more than did those steers grazing HE tall fescue pastures during the fall + winter grazing season, when evaluated with and without the GD covariate, respectively (Table 15). Steers grazing EFCL tall fescue gained 280 (with GD) and 286 (without GD) g d^{-1} more than did those steers grazing HECL tall fescue. As in the spring and spring+summer analyses, the effect of MECL was intermediate between those of EFCL and HECL (Table 16). Mean ADG estimates of steers grazing HECL and HE pastures were similar. Steers grazing EF tall fescue had significantly larger daily gains than did those steers grazing ME tall fescue (Table 16). On the other hand, mean ADG estimates of steers grazing ME and HE tall fescues were similar. As in the summer, steers grazing EF and EFCL pastures had similar daily gains estimates.

Effect of the Initial Weight and Steer Grazing Days Per Hectare Covariates on Fall+Winter ADG Estimates

The IW covariate explained a substantial portion of the residual variation in fall+winter ADG for the mixed model analyses with (α = 0.0055) or without (α = 0.0037) the GD covariate. Mean ADG estimates were adjusted upwards for pasture treatments which were grazed by steers having IW greater than the sample IW mean; the reverse occurred when steer IW was smaller than the sample IW mean (Table 17). The GD covariate did not remove a significant portion of the residual variation in

Table 15. Fall+winter ADG generalized least squares means for the treatments, estimated from models with and without the steer grazing days ha^{-1} (GD) covariate.

Tall Fescue Treatment	With GD	SE[+]	No GD	SE[+]	n[§]
	--------------- g steer d		----------------		
Endophyte-free	712	64	710	63	32
Moderately infested	547	76	547	75	11
Highly infested	520	60	499	56	27
Endophyte-free + clover	743	76	735	74	20
Moderately infested + clover	605	86	597	85	6
Highly infested + clover	463	63	449	61	28

[+] Broad inference space of different tall fescue pastures of similar endophyte infestation and future years within Tennessee, southwestern Alabama, and northern Virginia.

[§] On a pasture basis with 3 steers or more per pasture.

Table 16. Linear contrasts for steer ADG during the fall+winter grazing seasons, analyzed with and without the steer grazing days ha^{-1} (GD) covariate.

Fall+winter Contrast[+]	Covariate inclusion	Estimated difference	SE[§]	α
		- g steer^{-1} d^{-1} -		
EF vs. ME	With GD	165	56	0.0040
	Without GD	163	54	0.0031
ME vs. HE	With GD	26	68	0.6976
	Without GD	48	63	0.4492
EFCL vs. MECL	With GD	138	69	0.0493
	Without GD	138	66	0.0414
MECL vs. HECL	With GD	142	69	0.0412
	Without GD	149	65	0.0260
EFCL vs. EF	With GD	31	85	0.7272
	Without GD	25	82	0.7601

[+] EF = endophyte-free, ME = moderately infested, and HE = highly infested; EFCL = endophyte-free + clover, MECL = moderately infested + clover, and HECL = highly infested + clover.

[§] Broad inference space of different tall fescue pastures of similar endophyte infestation and future years within Tennessee, southwestern Alabama, and northern Virginia.

Table 17. Initial weights and steer grazing days ha^{-1} (GD) arithmetic means and standard errors (SE) for the tall fescue treatments during fall+winter.

Treatment	Initial Weight	SE	GD	SE
	-- kg --		-- d ha^{-1} --	
Endophyte-free	258.1	4.91	242.4	10.7
Moderately infested	242.6	8.48	250.2	15.7
Highly infested	264.9	3.37	291.6	15.8
Endophyte-free + clover	269.7	2.25	144.9	1.3
Moderately infested + clover	274.0	2.53	145.6	17.8
Highly infested + clover	267.7	1.78	212.6	2.7

fall + winter ADG (α = 0.2815) probably because stocking rate was usually relatively low and constant in fall + winter.

Relationship Between ADG and Tall Fescue Endophyte Infestation Levels

Current Knowledge

In order to establish some association between endophyte incidence and animal performance, most grazing trials have compared highly infested tall fescue to endophyte-free tall fescue. Most of these studies have found a strong linear relationship between endophyte level in tall fescue stands and reduction in steer gains during spring + summer, but not during fall (Williams et al., 1984; Stuedemann et al., 1985).

Crawford et al. (1989) regressed steer performance over a wide range of endophyte infestation levels and obtained a similar response to that obtained by researchers who compared only highly infested tall fescue with endophyte-free tall fescue. Fribourg et al. (1991b) found a curvilinear relationship between endophyte levels in tall fescue-clover mixtures and steer performance at moderate infestation levels in spring and spring + summer. These researchers determined that there was little difference in performance when steers grazed tall fescue-clover mixtures with 35 to 80% E+, but that the effect of 22% E+ was intermediate between those of 3% and 35% E+.

Non-equally Spaced Orthogonal Polynomial Contrasts Used to Estimate the Linear and Quadratic Effects of Endophyte Infestation Levels on Steer ADG

Non-equally spaced orthogonal polynomial contrasts of the GLS treatment means indicated that a strong linear relationship existed in all seasons between ADG and endophyte incidence in pure tall fescue stands and tall fescue-clover mixtures (Figures 2 to 9; Table 18). The quadratic effect was significant ($\alpha < 0.1029$) for steers grazing tall fescue stands of grass only in fall+winter (Table 18). On the other hand, the quadratic effect was not significant ($\alpha \geq 0.2240$) in the three other seasonal analyses, or in the fall+winter with clover analysis. Therefore, combining datasets from several independent grazing trials does indicate that a strong linear relationship exists between reduction in daily gains and increasing endophyte levels in all seasons, regardless of the presence of clover. This statement does not imply that the relationship is definitely linear in all discrete cases.

In the 1980's, several persons postulated that there was about a 45 g d^{-1} reduction in ADG for each 10% increase in endophyte infestation level.

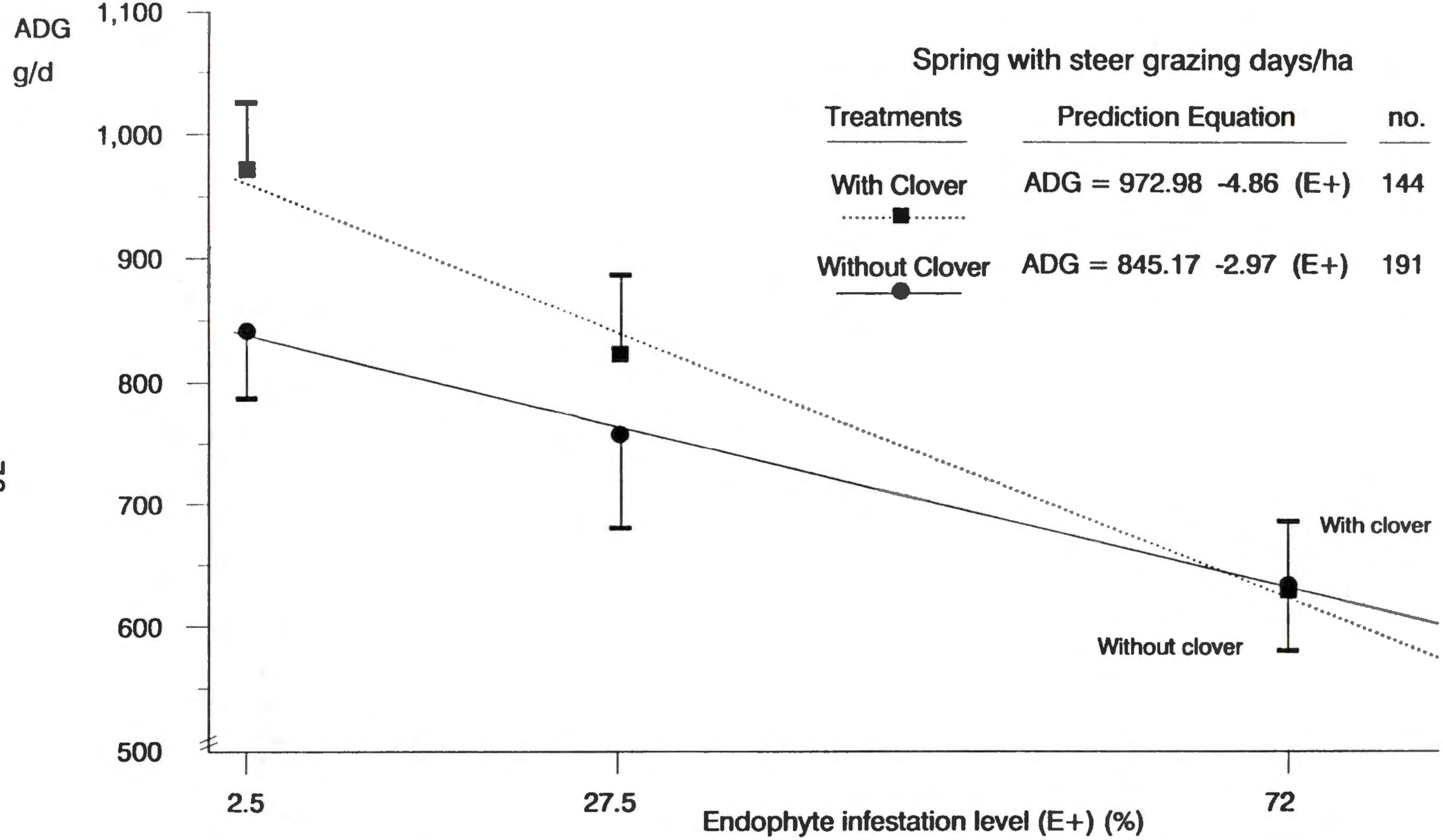

Figure 2. Spring ADG generalized least squares means of steers grazing tall fescue pastures, as a function of different levels of endophyte infestation, for mixed models including the covariate steer grazing days/ha.

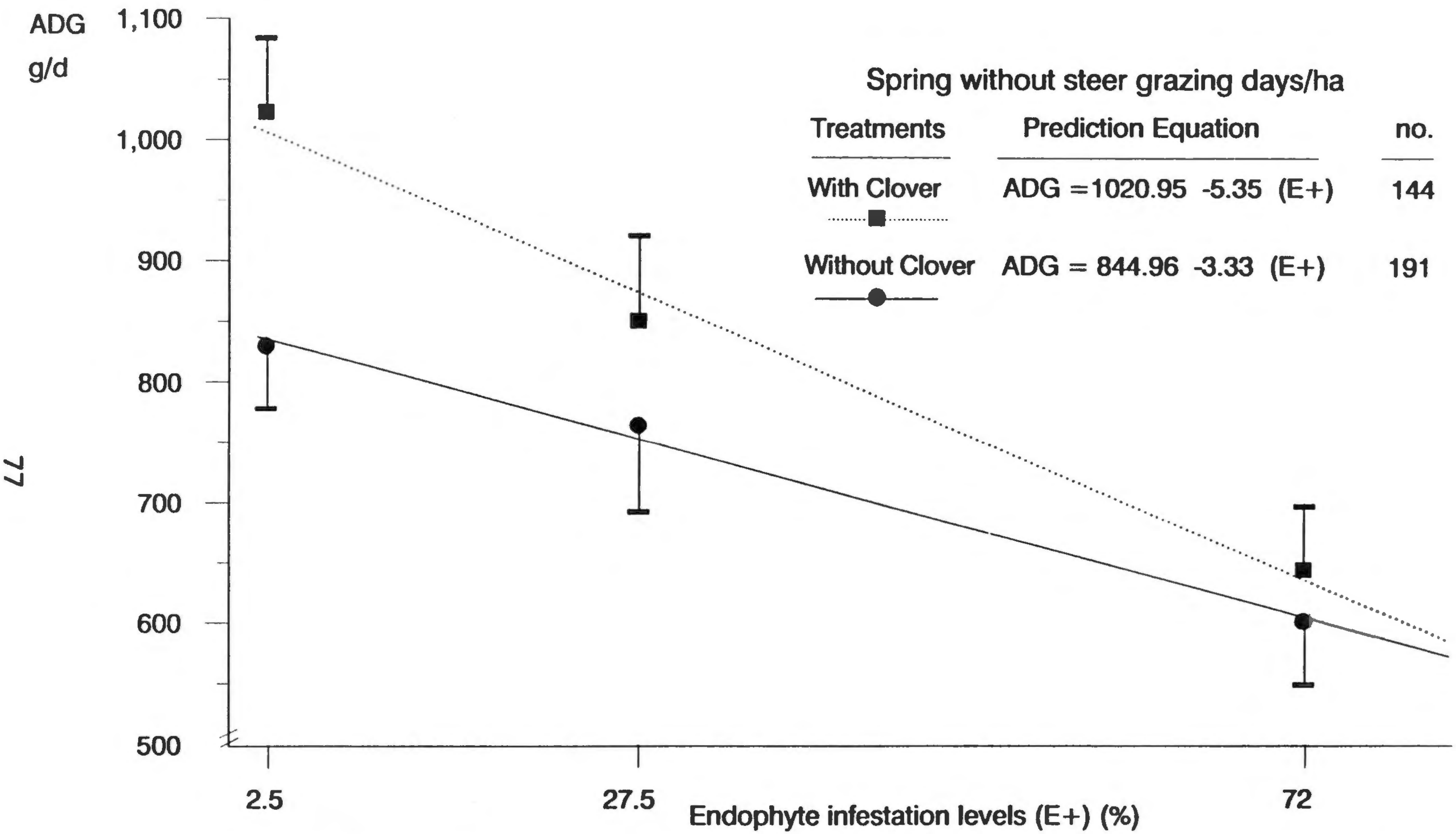

Figure 3. Spring ADG generalized least squares means of steers grazing tall fescue pastures, as a function of different levels of endophyte infestation, for mixed models excluding the covariate steer grazing days/ha.

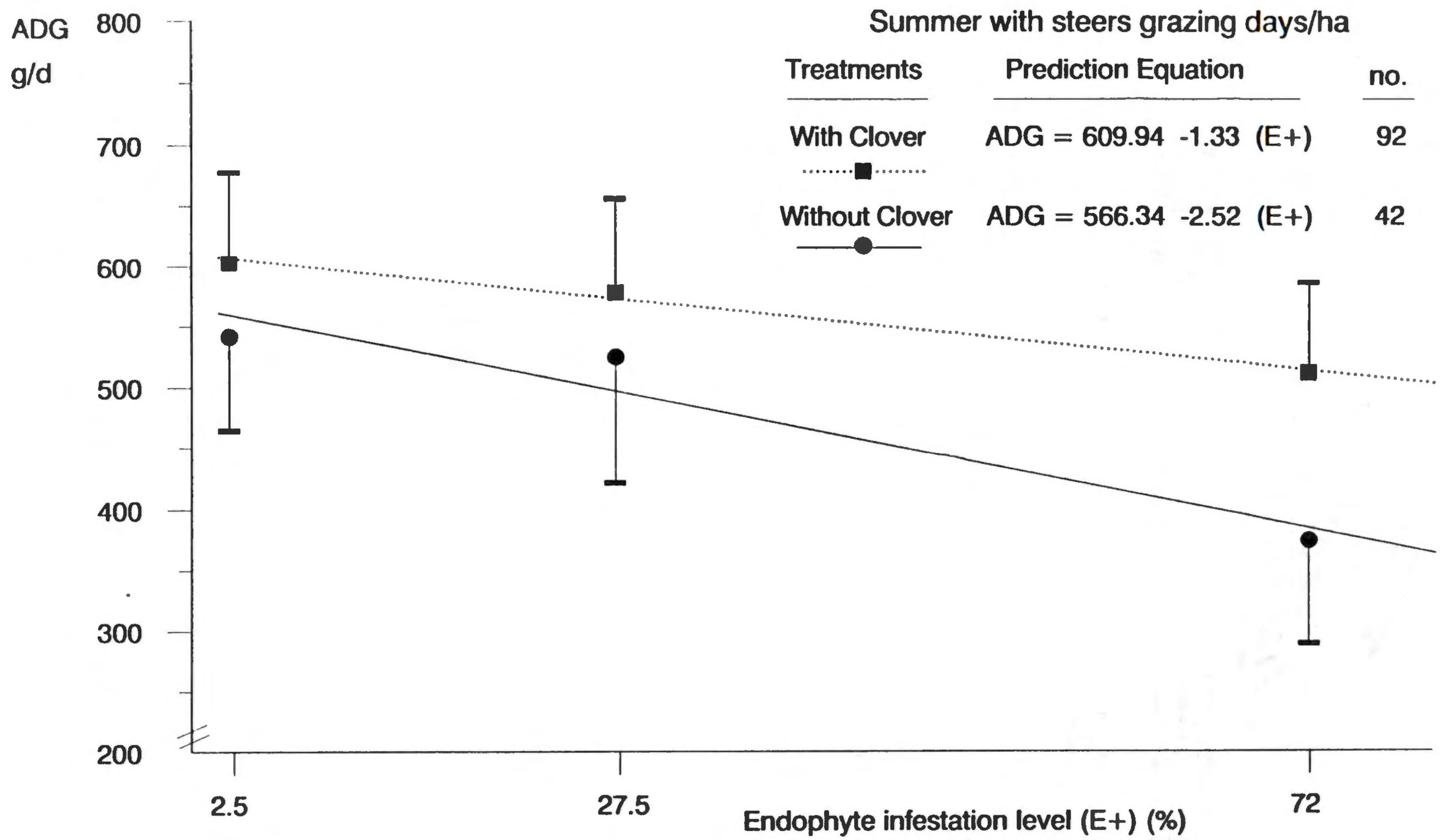

Figure 4. Summer ADG generalized least squares means of steers grazing tall fescue pastures, as a function of different levels of endophyte infestation, for mixed models including the covariate steer grazing days/ha.

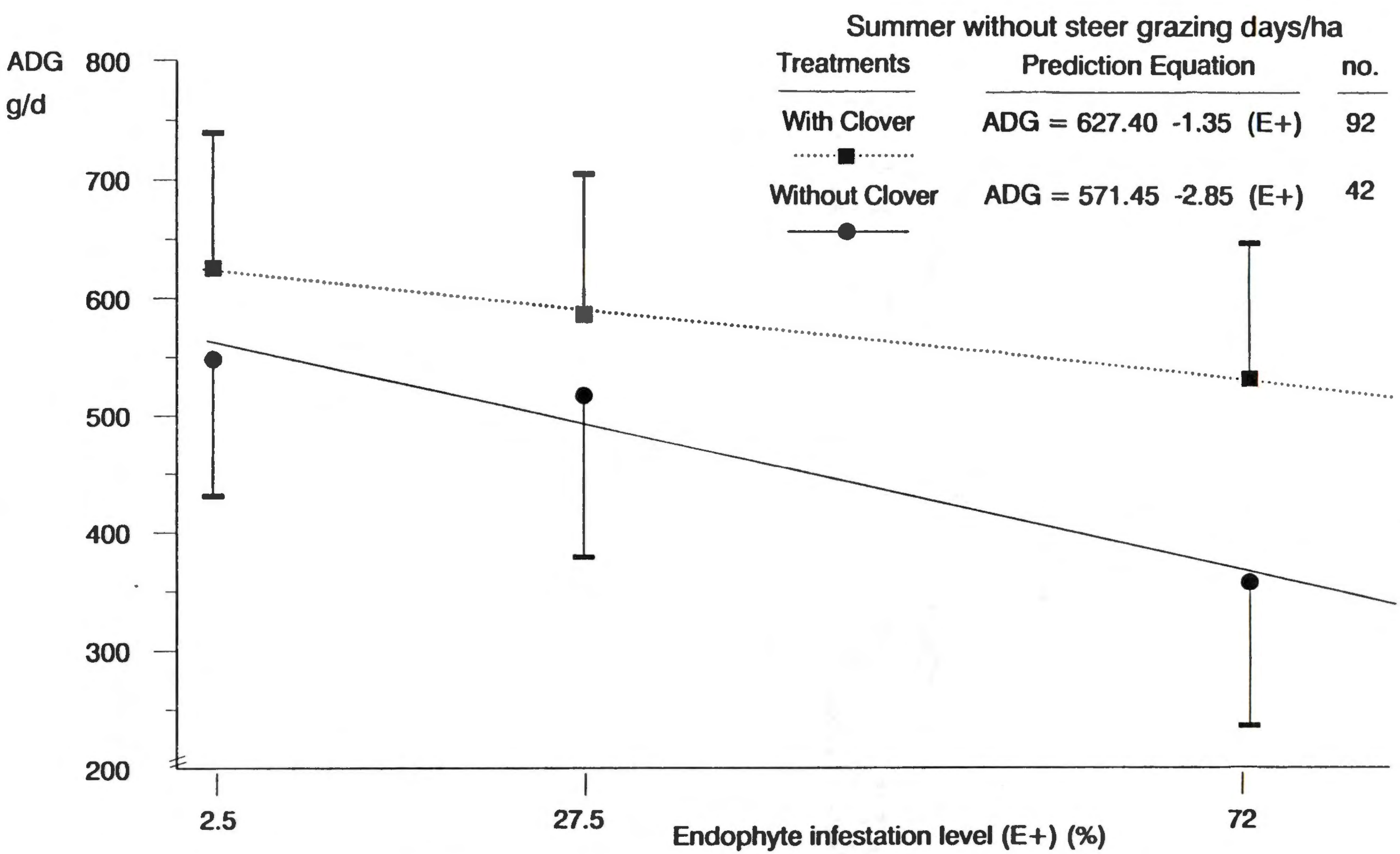

Figure 5. Summer ADG generalized least squares means of steers grazing tall fescue pastures, as a function of different levels of endophyte infestation, for mixed models excluding the covariate steer grazing days/ha.

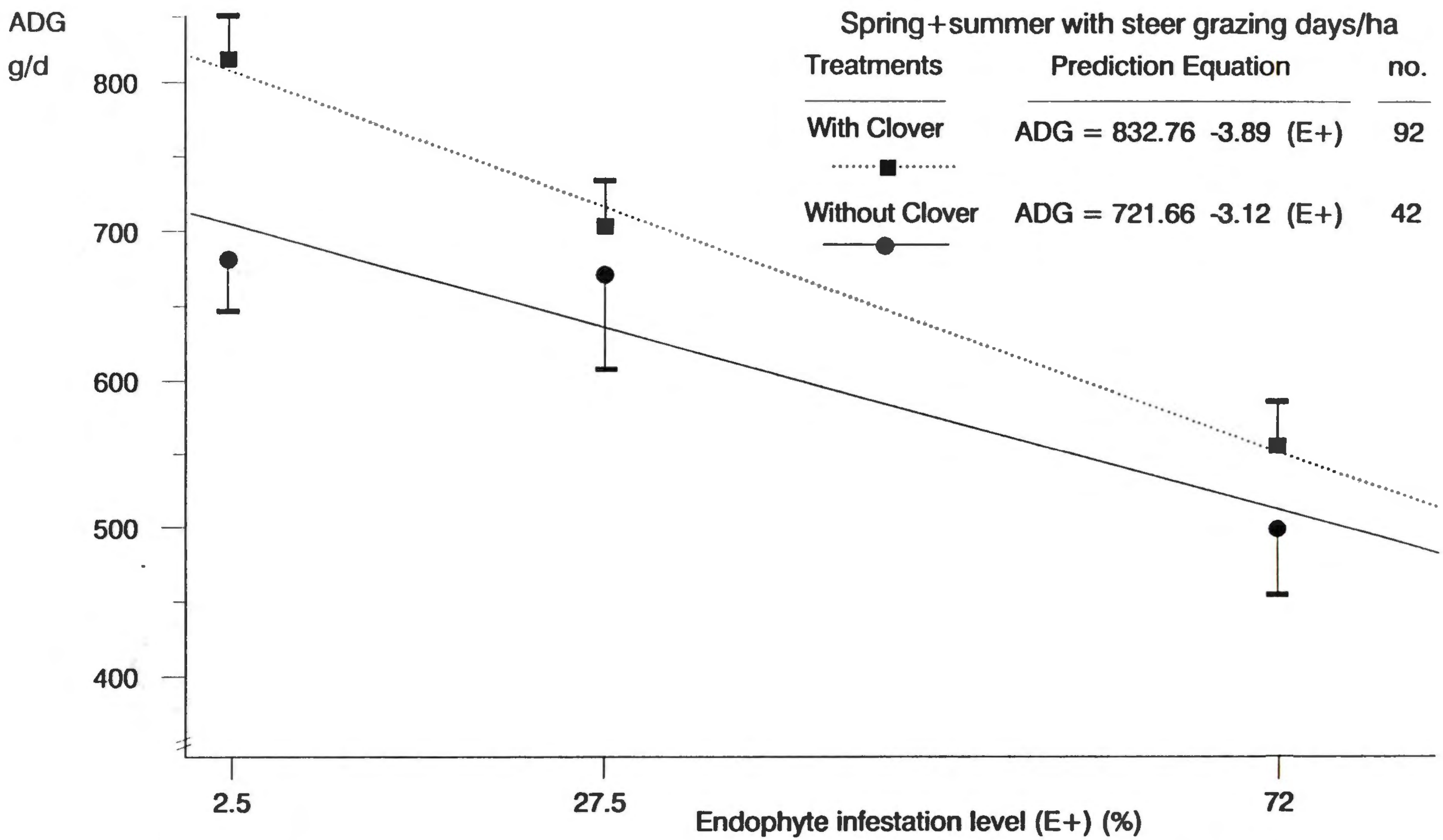

Figure 6. Spring+summer ADG generalized least squares means of steers grazing tall fescue pastures, as a function of different levels of endophyte infestation, for mixed models including the covariate steer grazing days/ha.

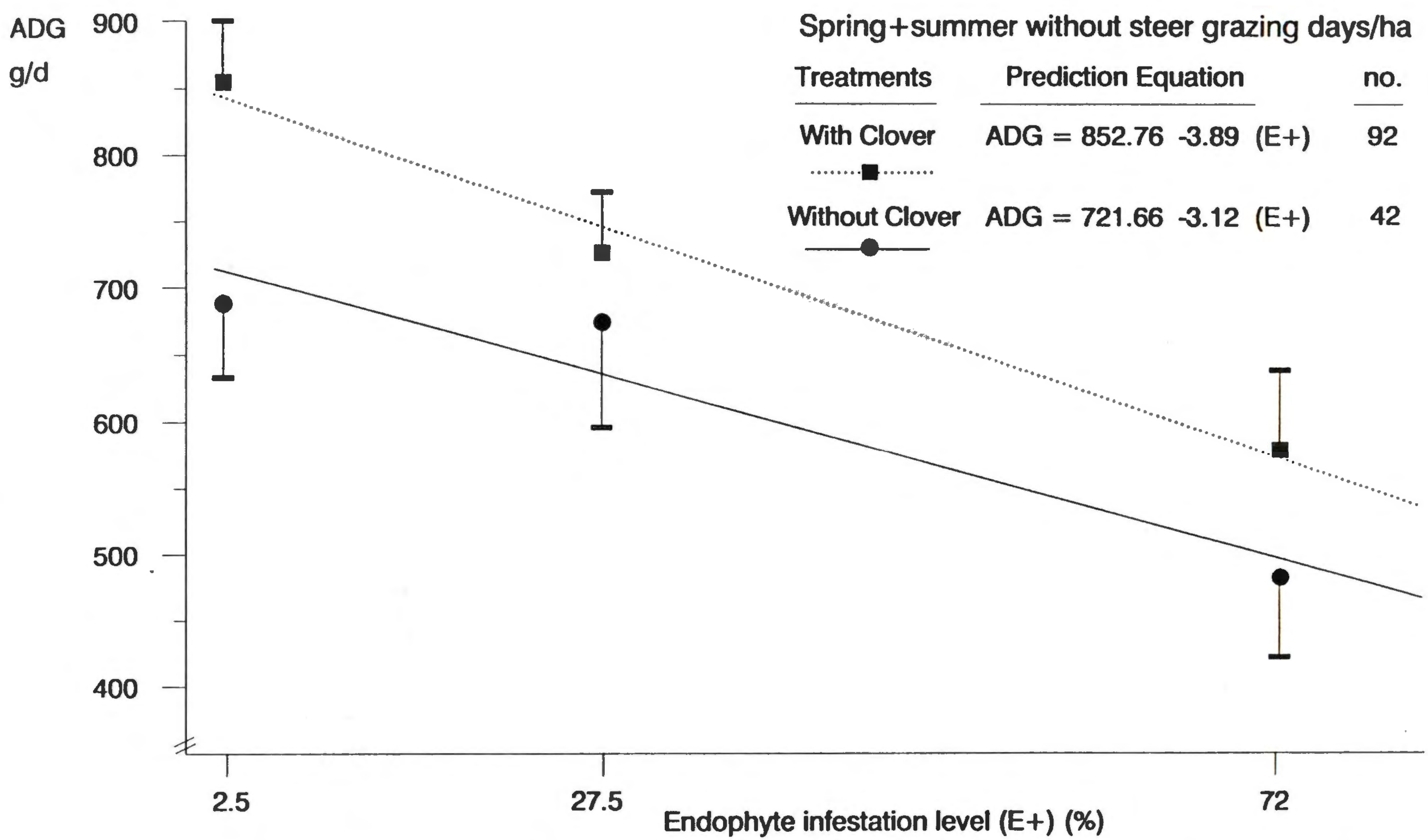

Figure 7. Spring+summer ADG generalized least squares means of steers grazing tall fescue pastures, as a function of different levels of endophyte infestation, for mixed models excluding the covariate steer grazing days/ha.

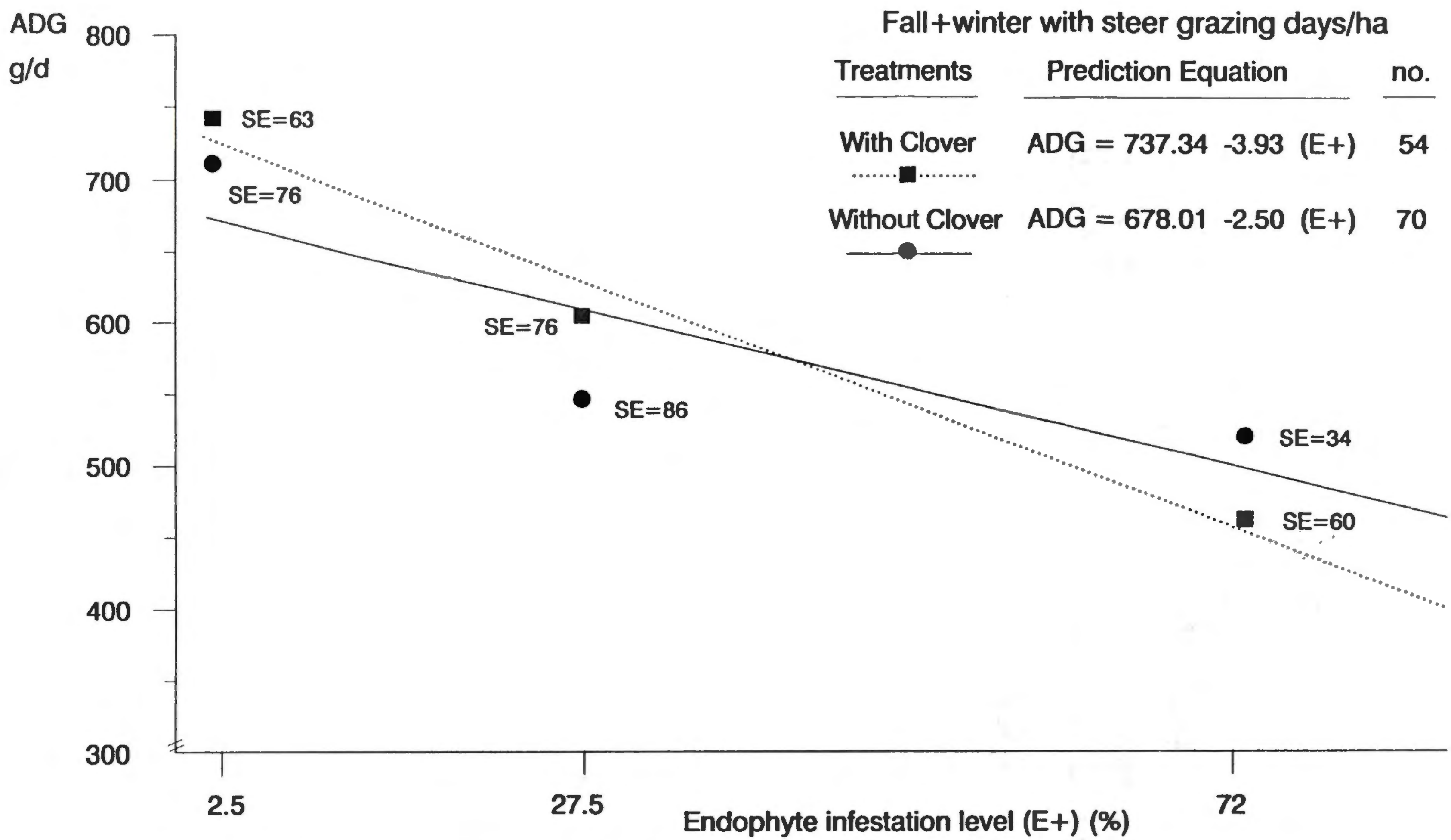

Figure 8. Fall+winter ADG generalized least squares means of steers grazing tall fescue pastures, as a function of different levels of endophyte infestation, for mixed models including the covariate steer grazing days/ha.

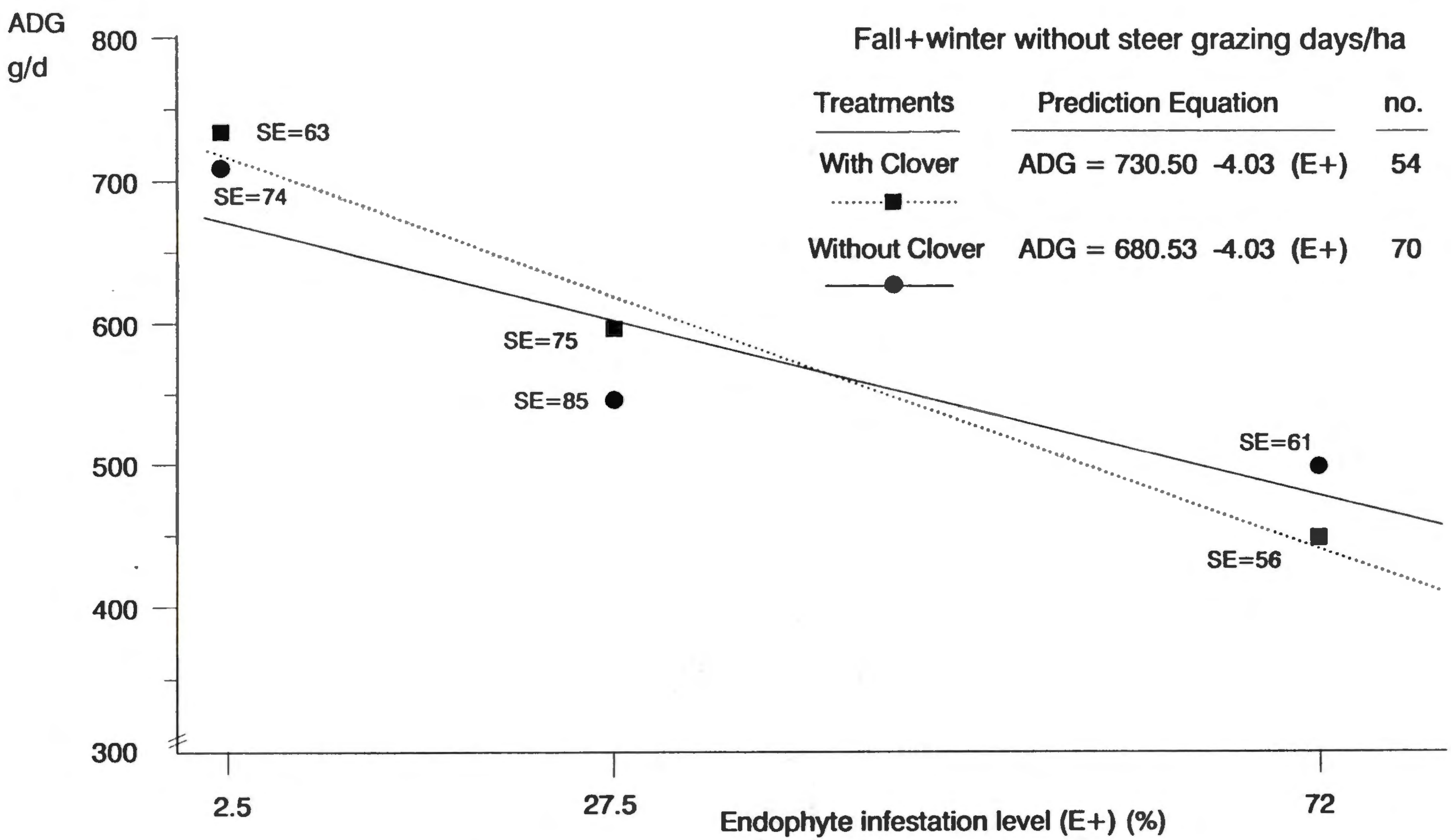

Figure 9. Fall+winter ADG generalized least squares means of steers grazing tall fescue pastures, as a function of different levels of endophyte infestation, for mixed models excluding the covariate steer grazing days/ha.

Table 18. Nonequally spaced orthogonal polynomial ADG contrasts for steers grazing tall fescue pastures free of the fungal endophyte or infested at two levels, with and without clover, during spring, spring + summer, summer, and fall + winter.

Polynomial Orthogonal Contrast⁺	Covariate inclusion§	F value	α
Spring			
Without Clover:			
Linear Effect	With GD	29.74	0.0001
	Without GD	33.61	0.0001
Quadratic Effect	With GD	0.12	0.7279
	Without GD	0.01	0.9459
With Clover:			
Linear Effect	With GD	39.50	0.0001
	Without GD	42.74	0.0001
Quadratic Effect	With GD	0.48	0.4684
	Without GD	0.70	0.4026
Spring + Summer			
Without Clover:			
Linear Effect	With GD	21.37	0.0001
	Without GD	24.20	0.0001
Quadratic Effect	With GD	0.73	0.3954
	Without GD	0.69	0.4095
With Clover:			
Linear Effect	With GD	84.77	0.0001
	Without GD	65.74	0.0001
Quadratic Effect	With GD	1.13	0.2893
	Without GD	1.50	0.2240

Table 18. (continued)

Polynomial Orthogonal Contrast+	Covariate inclusion§	F value	α
Summer			
Without Clover:			
Linear Effect	With GD	10.11	0.0020
	Without GD	10.30	0.0019
Quadratic Effect	With GD	0.22	0.6373
	Without GD	0.12	0.7272
With Clover:			
Linear Effect	With GD	5.71	0.0188
	Without GD	3.68	0.0585
Quadratic Effect	With GD	0.01	0.9951
	Without GD	0.08	0.7785
Fall + Winter			
Without Clover:			
Linear Effect	With GD	10.67	0.0016
	Without GD	16.47	0.0001
Quadratic Effect	With GD	3.03	0.0851
	Without GD	2.72	0.1028
With Clover:			
Linear Effect	With GD	24.68	0.0001
	Without GD	29.62	0.0001
Quadratic Effect	With GD	0.34	0.5602
	Without GD	0.32	0.5739

+ Mean levels of endophyte infestation of the endophyte-free, moderate and highly infested tall fescue treatments used to obtain the linear and quadratic coefficients, were 2.5, 27.5, and 74%, respectively.

§ For mixed models analyzed with and without the steer grazing days ha^{-1} covariate (GD).

Simple linear regression analyses of the GLS treatment means suggest that the reduction in ADG for each 10% increase in E+ infestation can vary considerably, depending primarily on the season and to a lesser extent on the presence of clover. The seasonal range of reduction in ADG was 53.5 to 13.3 g d^{-1} for steers grazing tall fescue-clover mixtures in spring and summer, respectively. The general rule-of-thumb appears to be more applicable to spring and spring+summer grazing than it is to summer grazing (Figures 2 to 7).

Crawford et al. (1989) and Stuedemann et al. (1986) indicated that there was not a significant linear reduction in daily gains with increasing E+ infestation in fall. In this study, the combined analyses did indicate that there was a highly significant (α = 0.0001) linear reduction in fall+winter ADG (Figures 8 and 9).

Generalized Least Squares Means of ADG for the Treatment X Location Combinations

Implied Inference Spaces for the Treatment X Location Combinations

In order to estimate the mean ADG for each treatment X location combination, all model effects were defined as random, except the main effects of locations and of treatments, and the interaction of locations and treatments. Since there was not a full ensemble of all treatments at each location, generalized least squares (GLS) means and their associated SE are specific for the location X treatment model effect. Therefore, mean ADG estimates and associated SE for these mixed model analyses represent the intermediate inference space of different tall fescue pastures during future years for these particular locations.

Variations in Average Daily Gains Among Combinations of Treatments and Locations

Generalized least squares mean estimates of ADG were variable for the treatment X location combinations during spring, summer, spring+summer, and fall+winter (Tables 19 to 22). The extreme to moderate variations in ADG means were expected because of the differences in environmental and climatic situations, *e.g.*, edaphic factors, available N, slopes, aspects, elevations, air temperatures, distribution and amounts of rainfall, lengths of growing season, *etcetera*. Comparability among different studies is also difficult, because of the problem of achieving complete objectivity in grazing research. However, the variations in environmental and climatic situations, and the slightly different methods and idiosyncrasies of investigators among locations, should be accepted as additional sources of true experimental error.

Table 19. Spring ADG generalized least squares means for Treatment X Location combinations, estimated from models with and without the steer grazing days ha^{-1} (GD) covariate.

Treatment	Location	With GD	SE+	No GD	SE+	n§
Endophyte-free:		-------------	g $steer^{-1}$ d^{-1}	--------------		
EF	Marion Junction, AL	890	117	868	123	24
EF	Hope, AR	889	196	867	205	2
EF	Calhoun, GA	885	148	856	155	12
EF	Mount Vernon, MO	975	137	1050	152	36
EF	Grand Junction, TN	981	95	807	96	12
EF	Knoxville, TN	763	160	768	169	16
Moderately endophyte infested:						
ME	Marion Junction, AL	706	117	694	123	9
ME	Knoxville, TN	767	170	771	180	4
Highly endophyte infested:						
HE	Marion Junction, AL	550	118	465	123	15
HE	Hope, AR	715	196	693	205	2
HE	Dixon Springs, IL	385	170	415	177	20
HE	Mount Vernon, MO	975	143	1050	144	9
HE	Grand Junction, TN	827	106	606	106	6
HE	Knoxville, TN	556	171	535	180	4
HE	Middleburg, VA	245	125	380	130	20
Endophyte-free + clover:						
EFCL	Hope, AR	1022	196	1001	205	2
EFCL	Calhoun, GA	999	145	1056	152	8
EFCL	Grand Junction, TN	1080	83	1014	87	36
Moderately endophyte infested + clover:						
MECL	Grand Junction, TN	893	87	820	92	12
MECL	Greeneville, TN	914	104	995	109	20
Highly endophyte infested + clover:						
HECL	Hope, AR	651	196	629	205	2
HECL	Dixon Springs, IL	435	173	534	181	10
HECL	Grand Junction, TN	744	87	612	90	24
HECL	Greeneville, TN	711	104	803	109	20
HECL	Middleburg, VA	274	122	411	127	10

+ Standard errors for the broad inference space of steer ADG during future years and different tall fescue pastures of similar botanical composition.

§ On a pasture basis with 3 steers or more per pasture.

Table 20. Summer ADG generalized least squares means for Treatment X Location combinations, estimated from models with and without the steer grazing days ha^{-1} (GD) covariate.

Treatment	Location	With GD	SE[+]	No GD	SE[+]	n[§]
		----------- g $steer^{-1}$ d^{-1}		---------		
Endophyte-free:						
EF	Calhoun, GA	634	159	727	130	2
EF	Grand Junction, TN	375	57	337	54	12
EF	Knoxville, TN	428	79	417	67	16
Moderately endophyte infested:						
ME	Knoxville, TN	391	100	361	91	4
Highly endophyte infested:						
HE	Grand Junction, TN	270	71	214	68	6
HE	Knoxville, TN	161	100	125	90	4
Endophyte-free + clover:						
EFCL	Calhoun, GA	799	146	894	112	4
EFCL	Grand Junction, TN	435	45	411	41	28
Moderately endophyte infested + clover:						
MECL	Grand Junction, TN	442	54	439	52	10
MECL	Greeneville, TN	772	54	797	52	16
Highly endophyte infested + clover:						
HECL	Grand Junction, TN	332	49	309	45	18
HECL	Greeneville, TN	728	54	753	47	16

[+] Standard errors for the broad inference space of steer ADG during future years and different tall fescue pastures of similar botanical composition.

[§] On a pasture basis with 3 steers or more per pasture.

Table 21. Spring+summer ADG generalized least squares means for Treatment X Location combinations, estimated from models with and without the steer grazing days ha^{-1} (GD) covariate.

Treatment	Location	With GD	SE[+]	No GD	SE[+]	n[§]
		----------- g $steer^{-1}$ d^{-1} -----------				
Endophyte-free:						
EF	Calhoun, GA	836	134	878	139	2
EF	Grand Junction, TN	678	49	631	53	10
EF	Knoxville, TN	544	70	546	82	16
Moderately endophyte infested:						
ME	Knoxville, TN	530	83	521	92	4
Highly endophyte infested:						
HE	Grand Junction, TN	530	59	465	61	6
HE	Knoxville, TN	301	83	284	92	4
Endophyte-free + clover:						
EFCL	Calhoun, GA	1034	126	1096	131	4
EFCL	Grand Junction, TN	799	38	792	45	28
Moderately endophyte infested + clover:						
MECL	Grand Junction, TN	684	44	679	50	10
MECL	Greeneville, TN	776	49	821	55	16
Highly endophyte infested + clover:						
HECL	Grand Junction, TN	525	42	508	47	18
HECL	Greeneville, TN	638	48	685	54	16

[+] Standard errors for the broad inference space of steer ADG during future years and different tall fescue pastures of similar botanical composition.

[§] On a pasture basis with 3 steers or more per pasture.

Table 22. Fall + winter ADG generalized least squares means for the Treatment X Location combinations, estimated from models with and without the steer grazing days ha^{-1} (GD) covariate.

Treatment	Location	With GD	SE+	No GD	SE+	n§
		---------- g $steer^{-1}$ d^{-1} ----------				
Endophyte-free:						
EF	Marion Junction, AL	765	88	768	84	24
EF	Knoxville, TN	387	196	378	182	8
Moderately endophyte infested:						
ME	Marion Junction, AL	591	104	595	98	9
ME	Knoxville, TN	305	209	294	195	2
Highly endophyte infested:						
HE	Marion Junction, AL	516	89	513	82	15
HE	Knoxville, TN	302	212	292	199	2
HE	Middleburg, VA	486	109	475	85	10
Endophyte-free + clover:						
EFCL	Grand Junction, TN	805	118	813	102	20
Moderately endophyte infested + clover:						
MECL	Grand Junction, TN	663	125	672	110	6
Highly endophyte infested + clover:						
HECL	Grand Junction, TN	529	118	537	102	18
HECL	Middleburg, VA	418	109	407	85	10

+ Standard errors for the broad inference space of steer ADG during future years and different tall fescue pastures of similar botanical composition.

§ On a pasture basis with 3 steers or more per pasture.

There are other tangible explanations for the moderate variability in ADG. Most of the variation among spring ADG estimates occurred for steers grazing highly infested tall fescue pastures, with (HECL) and without (HE) clover. There was less variability in spring ADG estimates among steers ingesting endophyte-free with (EFCL) and without (EF) clover, or moderately infested tall fescue, with (MECL) and without (ME) clover. For the model analyzed with the GD covariate, spring ADG estimates for steers grazing HECL and HE tall fescues ranged from 274 to 744 and from 245 to 975 g d^{-1}, respectively (Table 19). For the model analyzed without the GD covariate, spring ADG estimates for steers grazing HECL and HE tall fescues ranged from 411 to 629 and from 380 to 1050 g d^{-1}, respectively. Therefore, the inconsistencies in spring ADG appear to be associated primarily with highly infested tall fescue pastures, regardless of the presence of clover.

It is probable that environmental and climatic variations among locations caused daily gains of steers grazing highly infested tall fescue to be different. Read and Camp (1986) reported that depressed steer performance due to endophyte infestation was not consistent among years. It may be influenced by environmental factors that either affect production of compounds responsible for tall fescue toxicosis or accentuate the physiological effects these compounds have on steers.

The genotypes of tall fescue/*A. coenophialum* complexes may have been different among locations. It is known that not all isolates of *A. coenophialum* produce the same amount of alkaloids (Bacon, 1988; Siegel et al., 1990). The alkaloid concentrations in the infested tall fescue pastures at each location in this study were not determined. It would be advantageous in future grazing trials to determine the kinds and concentrations of specific alkaloids occurring in the infested tall fescue pastures or in the consuming animals (Savary et al., 1990).

Most researchers try to obtain a uniform group of experimental steers, often resulting in a gene pool of common sires within a grazing trial. It is possible that a particular line(s) of steers may be more tolerant to tall fescue toxicosis than are other lines. Cattle purchased from outside of the southeast US, which have not had an endophyte infested tall fescue diet, tend to exhibit more severe signs of tall fescue toxicosis than do cattle that have grazed infested tall fescue exclusively (John C. Waller, personal communication). Some researchers and/or producers may cull more effectively those animals which exhibit signs of tall fescue toxicosis than do others. These suppositions may explain partially why there is a moderate degree of non-uniformity among locations using similar infested tall fescue pasture treatments.

The smallest spring ADG estimates for steers grazing HECL and HE tall fescues were obtained from the study conducted at Middleburg, VA. These

ADG estimates were for steers grazing during the early spring. These same steers also had grazed tall fescue throughout the preceding winter. Spring ADG estimates probably would have been larger at this location if grazing had continued at least into May, allowing steers to overcome the previous effects of winter grazing. At this location, the fall+winter daily gains were similar to those obtained at other locations (Table 23).

Spring ADG estimates of steers grazing HECL and HE tall fescues were also small for the study at Dixon Springs, IL (Table 19). The mean stocking rate of the HE tall fescue pastures at this location was much higher than the mean stocking rate at the other locations (6.9 versus 5.7 steers ha^{-1}, respectively). The mean stocking rate of the HECL tall fescue pastures at Dixon Springs, IL was slightly higher than the mean stocking rate for all other locations (5.2 versus 4.8 steers ha^{-1}, respectively).

The mean spring ADG estimates of steers grazing HE tall fescue was exceptionally large in the study conducted at Mount Vernon, MO. Spring ADG means at this location were 975 and 1050 g d^{-1}, when estimated from models with and without the GD covariate, respectively. These large gains can be attributed to the short durations of spring grazing (35 to 56 d), which may not have allowed sufficient time for the development of strong tall fescue toxicosis signs in test steers.

Researchers at Mount Vernon, MO remove steers from endophyte infested tall fescue pastures in late May to early June. Summer grazing consists of other cool-season and warm-season grasses after spring tall fescue use. Several years of past poor animal performance obtained from infested tall fescue during the late spring and summer led to this management change (Richard J. Crawford, personal communication). At some locations, producers may benefit from such a practice or from having an endophyte-free tall fescue pasture for late spring and summer grazing.

Adjustment of the Treatment X Location Means for the Initial Weight and Steer Grazing Days per Hectare Covariates

For mixed models analyzed without the GD covariate, the IW covariate removed a significant portion of the residual variation in ADG for the summer (α = 0.0250) and fall+winter (α = 0.0314) analyses. Treatment X location estimates were adjusted upwards for pastures stocked with steers having IW greater than the sample IW mean, a biologically rational adjustment; the reverse occurred when location X treatment IW were smaller than the sample IW mean. The IW covariate was not significant (α > 0.15) for the spring and spring+summer analyses.

The GD covariate removed a significant portion of the residual ADG variation in each spring (α = 0.0001), summer (α = 0.0104), and spring+summer (α = 0.0054) mixed model analysis. Mean ADG estimates were adjusted upwards for treatment x location combinations that had a larger number of GD than the sample GD mean -- this upward adjustment is also biologically rational. The sample GD means for spring, summer, spring+summer, and fall+winter seasons were 414, 288, 805, and 224 d ha^{-1}, respectively. The GD covariate did not explain additional variation in steer ADG for the fall+winter analysis (α = 0.8168), indicating that tall fescue pastures were stocked uniformly, and that the steers remained on pasture for similar durations during this period.

The differences between ADG mean estimates evaluated with and without the GD covariate were usually negligible. The small differences between the ADG means should not be considered as an inability of the GD covariate to explain a significant proportion of the residual error, because the probability values for GD were highly significant for spring, summer, and spring+summer, but not for the fall+winter.

The small differences between ADG means analyzed with and without the GD covariate simply demonstrate the ability of Henderson's mixed models procedure to distribute the realized values of the random effects depending on the model specified. For those models which did not include the GD covariate, the variation was partitioned into additional random effects that were negligible in the models which included the GD covariate, or it inflated the variance components for random effects that were significant for the model containing GD (Table 23). For example, the variance components for the random effects of Years(Locations) and

Table 23. Magnitude of the maximum likelihood (ML) variance components for mixed models, with and without the steer grazing days ha^{-1} (GD) covariate, used to estimate mean ADG for treatment X location combinations during spring, spring+summer, summer, and fall+winter.

Season	Random effect+	ML Variance Components§ With GD	No GD
Spring	Year (Location)	42197	47887
	Block (Year X Location)	12272	12220
	Residual Error	18559	20621
Spring+ Summer	Year (Location)	0	4168
	Treatment X Year (Location)	0	1032
	Block (Year X Location)	12153	9048
	Residual Error	9693	8396
Summer	Year (Location)	0	849
	Treatment X Year (Location)	0	1276
	Block (Year X Location)	14095	4993
	Residual Error	17244	17766
Fall+ Winter	Year (Location)	30425	27494
	Treatment X Year (Location)	2052	1359
	Residual Error	14002	14064

+ The full mathematical model was: ADG = Location + Treatment + [Location X Treatment] + Year (Location) + [Treatment X Year(Location)] + [Block(Year X Location)] + Initial Weight + with or without steer grazing days ha^{-1} + ϵ. All effects were random except the main effects of treatments and locations, and the interaction of treatments X locations.

§ Seasonal ADG variance components not listed from the full model were zero for mixed models with and without the steer grazing days ha^{-1} covariate.

[Treatments X Years(Locations)] were null for mixed models which included the GD covariate during spring+summer and summer. In contrast, the variance components for these same random effects were large for mixed models analyzed without the GD covariate.

The net result was that residual variance components were similar for both analyses because the models without the GD covariate distributed random variance into additional random effects, or they inflated certain effects that could be correlated with variability in GD. This situation did not occur for the fall+winter analyses because the pasture stocking rates and grazing durations were similar. Partitioning random variation could not have been accomplished as effectively if the studies had been analyzed independently because replication over time was restricted usually from 2 to 3 years and replication over space (locations) was naturally absent.

Summary Discussion on the Treatment X Location ADG Means

Treatment x location combinations data indicate that daily gains will be variable when steers graze highly infested tall fescue, suggesting that the error structure is very complex. Additional studies are needed to evaluate quantitatively the effects of temperature and humidity, edaphic conditions, alkaloid concentrations found in tall fescue and the rumen, intake levels, *etcetera* on steer ADG. Knowledge of the effect(s) that these variables have on steer ADG may allow for more definitive conclusions about the variability among highly infested tall fescue treatments within the transition zone. Daily gains were generally poor for steers grazing highly infested tall fescue, regardless of the presence of clover. Nonetheless, it is obvious that some highly infested tall fescue pastures will provide above average steer performance during some favorable years (climates) and at certain locations (environments) while others will not. Excellent performance was obtained for steers grazing endophyte-free tall fescue pastures, with and without clover, at all locations represented in the study. Mean ADG for steers grazing the moderately infested tall fescue, with and without clover, was usually intermediate between those for steers grazing endophyte-free or highly infested tall fescues.

Use of the Combined Variance Components in Future Grazing Trials

The seasonal variance components could be used in future tall fescue grazing trials as known sources of variation (Table 24). This may allow the researcher to forgo replication over time and space and in turn investigate a larger number of treatments, which might include two or more stocking rates. In this sense, the researcher could take advantage of information external to his/her experiment. Why depend on variances estimated from

Table 24. Magnitude of the maximum likelihood (ML) variance components for mixed models, with and without the steer grazing days ha^{-1} (GD) covariate, used to estimate mean ADG in g d^{-1} for the treatments during spring, spring+summer, summer, and fall+winter.

Season	Random effect⁺	ML Variance Components[§£] With GD	No GD
Spring	Year (Location)	59215	63859
	Location X Treatment	1797	1992
	Treatment X Year (Location)	1397	2057
	Block (Year X Location)	12477	12414
	Residual Error	18824	20710
Spring+ Summer	Location	0	4443
	Year (Location)	1861	8159
	Treatment X Year (Location)	0	1032
	Block (Year X Location)	12302	9349
	Residual Error	9796	8382
Summer	Location	15268	32109
	Year (Location)	996	4250
	Treatment X Year (Location)	0	2031
	Block (Year X Location)	17485	53898
	Residual Error	17606	17567
Fall+ Winter	Year (Location)	31229	31107
	Treatment X Year (Location)	2460	1892
	Residual Error	14051	14116

⁺ The full mathematical model was: ADG = Location + Treatment + [Location X Treatment] + Year (Location) + [Treatment X Year(Location)] + [Block(Year X Location)] + Initial Weight + with or without steer grazing days ha^{-1} + ϵ. All effects were random except the main effect of treatments.

§ Seasonal ADG variance components not listed from the full model were zero for mixed models with and without the covariate steer grazing days ha^{-1}.

£ ADG variance components are not unit dependent, *e.g.*, to reflect ADG in terms of lb d^{-1}: 1) divide each variance component by 1000^2 to express ADG in kg d^{-1}, and 2) multiply the quotient by 2.202^2 to express ADG in lb d^{-1}.

a single tall fescue grazing trial based on few degrees of freedom when combined variance components are available? The use of these known variance components may also be the best realistic way to avoid logistical and financial constraints that force compromises in the sizes of grazing trials.

These variance components may also be of value to modelers of forage-livestock systems. Systems analysis has become a useful tool for examining the forage-livestock interface. These components of variation could provide modelers with the coefficients needed to simulate data which epitomize the broad inference space.

SUMMARY

Most information concerning the effects of the tall fescue endophytic fungus *A. coenophialum* on livestock performance has been obtained from grazing trials conducted as discrete endeavors. These trials may be related over space and time. Henderson's mixed model procedure (MMP) provides the opportunity to combine several related studies in order to estimate steer performance over a broad inference space of future grazing years and different tall fescue pastures of similar endophyte infestation. The MMP allows the incorporation of the variance components of the random effects of years, locations, pastures, and blocks, and all random-random and random-fixed interactions into the mixed model equations, leading to generalized least squares estimates of the fixed treatment effects. A combined analysis of steer performance on tall fescue pastures, with and without clover, was done using datasets from several grazing studies conducted during the last 13 years at nine locations in seven eastern states. Steers grazing endophyte-free (EF) and moderately infected (ME $\geq$ 20% to $\leq$ 35% E+) tall fescues, with or without clover, had greater ADG than did those steers grazing highly infested (HE $\geq$ 50% to $\leq$ 97% E+) tall fescue, when estimated from models with and without the steer grazing days ha^{-1} covariate. The inclusion of clover into EF and ME tall fescue pastures improved steer daily gains. On the other hand, daily gains of steers grazing EF plus clover mixtures were usually greater than those of steers grazing ME plus clover mixtures. The inclusion of clover into HE tall fescue stands improved summer ADG only. Steers grazing EF plus clover mixtures had larger daily gains than those steers grazing grass-only EF stands in spring and spring+summer, but they did not in summer or fall+winter.

Seasonal, non-equally spaced polynomial contrasts of the generalized least squares treatment means indicated that a strong linear relationship existed in all seasons between ADG and endophyte incidence in tall fescue stands and tall fescue-clover mixtures. The combined analysis suggests that the reduction in ADG for each 10% increase in endophyte infestation can vary considerably, depending mostly on the season of the year and to a lesser extent on the presence of clover.

Mean ADG estimates were variable for the treatment X location combinations, but most of the variation occurred within the HE and HE plus clover treatments, suggesting that the error structure is very complex. Variable climatic and environmental situations probably caused much of the non-uniformity among locations. The genotypic variations among infested tall fescues, endophytic fungus strains, and consuming steers may also have contributed to the variability in ADG estimates among locations. Nonetheless, it is apparent that some producers will obtain satisfactory steer performance from highly infested stands during some favorable years (climates) and locations (environments), while others will not.

Pooling datasets provided a larger spectrum of years, pastures, and hence environments, than had been available for analysis at individual locations. Consequently, more precise estimates of steer performance subjected to a larger number of tall fescue treatments were obtained. The results from the combined analysis also provided a more coherent body of information than did the results obtained from each discrete trial, because the treatment means were adjusted for the initial steer weights and for steer grazing days ha^{-1} covariates. Combining datasets may be a feasible and realistic way to avoid logistical and financial constraints that force undesirable compromises in the conduct of grazing and other expensive or time-consuming research. The establishment of cooperative projects, using common treatments and identical protocols, would further increase the sensitivity of combined analyses.

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