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ASSESSING PERSISTENCE, PRODUCTIVITY, ANIMAL PREFERENCE, FORAGE QUALITY AND BOTANICAL COMPOSITION OF ROTATIONALLY GRAZED PERENNIAL COOL SEASON GRASSES AND CLOVERES GROWN IN MONOCULTURE AND BINARY MIXTURES IN MICHIGAN

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ABSTRACT

ASSESSING PERSISTENCE, PRODUCTIVITY, ANIMAL PREFERENCE, FORAGE QUALITY AND BOTANICAL COMPOSITION OF ROTATIONALLY GRAZED PERENNIAL COOL SEASON GRASSES AND CLOVERS GROWN IN MONOCULTURE AND BINARY MIXTURES IN MICHIGAN

By

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Most perennial cool season grasses and clovers introduced to Michigan have not been evaluated in binary mixtures. The objectives of this study, which was conducted over five growing seasons at three different latitudes in Michigan were: (i) to determine persistence and productivity of perennial grass and clover species grown in monoculture and binary mixtures under three different latitudes in Michigan; (ii) to determine botanical composition of grass-clover binary mixtures over time ; (iii) to determine animal preference and forage quality of these species under rotational grazing, and (iv) to evaluate the use of Near-Infrared Reflectance Spectroscopy (NIRS) to predict the species composition using three calibration equations. Grass-clover binary mixtures resulted in greater persistence, increased ground cover, greater animal preference and higher forage quality than grass or clover growing in monoculture. However, persistence of both perennial grass and clover was related to the site of the study. Kura clover provided the highest dry matter (DM) yield (tones acre^{-1}) and exceptional persistence under grazing particularly at southern part of Michigan. NIRS had higher prediction accuracy with R^2 ranging from 0.67 to 0.72 and SEP from 6.9 to 12.8 respectively using equations generated from hand separations of several grass and clover species. NIRS can be used to replace the hand separation method to determine botanical composition pasture species using a calibration equation developed from a large data set of hand-separated samples.

DEDICATION

To My Parents, My Wife And My Children

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GENERAL INTRODUCTION

Forage crops, by definition, are crops grown primary for feeding livestock. Forage crops can be harvested by machines or grazed by animals. Grazed forage is defined as forage, which is consumed directly by the grazing animals (Pasturage) (Vallentine, 2001). Pasture refers to the fenced area of domesticated forages on which animals are grazed. In Michigan more than 1.5 million hectares of farmland is dedicated to forage crops used for hay, silage, green chop, and over 500 thousand hectares are pastureland (2002 Census of Agriculture).

The goal of pasture management is to maximize forage quality and yield, while ensuring stand persistence. Stand persistence depends in part on management practices such as controlling weeds, maintaining proper soil fertility, and using an appropriate harvest or grazing schedule, but it also depends on the innate compatibility and winter hardiness of the forage species or cultivars used in pastures.

Severe winter conditions can cause damage to perennial forage crops in northern latitudes (Bèlanger et al., 2002). Introducing a new species requires knowledge of the species tolerance to local or regional environmental conditions. Pasture managers in northern temperate climates could improve their production and lengthen stand life by selecting high yielding winter hardy cultivars for pasture production. Since most pastures are planted with both legume and perennial grass, it is important that species selection is based on their performance in binary mixtures. However, there is limited commercial information available on pasture species performance in binary mixtures because seed companies mainly test their species in a monoculture.

Plant performance in pastures cannot be understood without reference to animals. Under grazing, animals tend to prefer the most palatable plants first, which consequently causes overgrazing of the most palatable species (Vallentine, 2001). Consumption of the more palatable species can also have an impact on the mixture composition. Hence, animal preference is a factor that may influence the relationship between legume and grasses in the pasture system. Forage nutritive evaluation provides the final assessment of pasture in terms of nutritional values, which ultimately can be vital in a pasture forage evaluation program.

The botanical composition need to be determined to estimate the percentage of actual legume and grass content in a pasture. Several methods have been used for estimating botanical composition of grass and legume mixtures. The most common method involves manually separating the mixtures into grass, legume, and weed components. However, this strategy is not practical since it is laborious, time-consuming, costly, and prone to operator error, especially when a large numbers of samples are being processed.

Near-Infrared Reflectance Spectroscopy (NIRS) can lead to more efficient determination of various components of forage quality. NIRS might be used successfully for analyzing forage components as a rapid and inexpensive technique to replace the hand separation approach. Historically, the first application of NIRS was reported in literature in 1939 (Gordy and Martin, 1939). The potential of NIRS for solving analytical problems was developed by Kay (1954). In 1968, Ben-Gera and Norris applied NIRS to analysis of agricultural products. NIRS was first shown by Norris et al. (1976) to be a rapid method to determine the chemical composition of forages.

This research was conducted to assess the persistence of grass and clover species and cultivars grown in monoculture and binary mixtures at three different latitudes in Michigan. Animal grazing preferences and forage quality were used in assessing the nutritive values and palatability of species and cultivars in this study. In addition, this study evaluated the use of NIRS for providing rapid and accurate results for predicting the botanical composition of grass and clover species.

This dissertation is divided into three studies. The objective of the first study, Chapter 1, was to assess the persistence and productivity of grass and clover cultivars grown in monoculture and binary mixtures established in three latitude locations in Michigan. Several variables were used for this evaluatation including: total forage yield, winter hardness, estimation of the grass and clover botanical composition, determining clover and grass ground cover and the percent clover in grass-clover mixtures over time.

The objective of the second study, Chapter 2, was to determine animal preferences and forage nutritive evaluation of perennial grass and clover species and cultivars established in monocultures and binary mixtures. The grazing trial was conducted in a single location. However, the nutritive evaluation was completed on all perennial grasses and clovers from three locations.

The objective of the third study, Chapter 3, was to determine whether NIRS can be applied as an alternative technique to the hand separation method for estimating grass-clover botanical composition. In this study, three calibration equations were created. The first equation was developed from pure samples obtained from a single location and year, which were mixed artificially to different proportions; the second equation was developed from hand separated samples collected from a single location and year and the third equation created from hand separated samples from three locations over three years with an additional constituent, weeds. All the three equations were developed to predict grass-clovers botanical composition collected from samples from three locations during 2003-2005 seasons. Calibration and validation equations were reported along with coefficients of determination (R^2) and standard errors of prediction (SEP).

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Chapter 1

EVALUATION OF THE PERSISTENCE AND PRODUCTIVITY OF PERENNIAL COOL SEASON GRASSES AND CLOVERS GROWN IN MONOCULTURE AND BINARY MIXTURES IN MICHIGAN

ABSTRACT

Persistence of grass-legume binary mixture components is important for pasture management, yet one species is often lost from the stand within a few growing seasons. Forage and livestock producers seek species and cultivars, which provide the greatest persistence and dry matter (DM) yield over the longest time. The objective of this study was to evaluate the persistence and DM yield of several grass and clover cultivars established in monoculture and binary mixtures across three locations representing different latitudes in Michigan. The evaluation criteria were based upon winter injury, percent ground cover, percentage clover in the grass-clover binary mixtures and total DM yield. Binary mixtures resulted in increased DM yield and resistance to winter injury compared to monoculture treatments. Even non-winter hardy grass species had higher persistence when associated with clovers compared with grass monoculture. Significant differences in performance among grass cultivars were observed with tall fescue shown significantly higher persistence and yield stability at all locations. Variations between latitude locations were observed. Among clover monoculture species, kura clover (Endura) had the greatest persistence and DM yield at the southern latitude in both monoculture and binary mixtures. This study demonstrated the importance of proper species and cultivars selection based upon location for persistence and dry matter production.

INTRODUCTION AND BACKGROUND

Several temperate forage grasses are commonly grown in binary mixtures with alfalfa (*Medicago sativa* L.), for hay or silage production (Casler, 1988). Most forage grasses grown in Michigan pastures can be classified as cool season plants (C3 plant), which have an optimum growth temperature of 18°-24 °C (Rohweder and Albrecht, 1995). During the mid to late summer, both heat and rainfall deficits often exceed the optimal growth range for cool season grasses, causing them to go into a drought-induced dormancy. During this period, there is often a shortage of soil moisture available for the shallow rooted cool season grasses (Penn State University Agronomy Guide, 2005-2006). Thus, many producers include legumes such as clovers in their pasture mixtures since the deeper taproots of clovers are able to utilize the soil moisture and minerals in the sub soil. Clovers species are often preferred for pasture over alfalfa as they are more tolerant to frequent grazing than alfalfa.

Forage grass-legume binary mixtures are important components of dairy and livestock diets, which offer many advantages over pure stands (Vough, et.al., 1995). Grasses provide a sod that can be helpful for increasing water infiltration and reducing runoff and soil erosion (Wadleight, et al., 1974; Wischmeier and Smith, 1978). Binary mixtures are more competitive with weeds than pure stands (Drolsom and Smith, 1976). Moreover, grasses benefit from fixing the atmosphere nitrogen (N) provided by legumes, which reduces the need for supplemental nitrogen fertilization (Vough, et al., 1995). Grass-legume binary mixtures may yield greater dry matter (DM) than legume monoculture (Chamblee and Collins, 1988). In contrast, alfalfa-grass binary mixtures did not yield more forage than an alfalfa monoculture at high seeding rates (Wilsie, 1949). In addition, legume-grass binary mixtures had higher in vitro DM digestibility (IVDMD), crude protein (CP) and better seasonal yield distribution than monoculture treatments. Binary mixed stands can reduce the risk of bloat, which is a very common problem with ruminants associated with grazing exclusively legume pastures (Casler and Walgenbach, 1990). In this study, we evaluated the persistence and DM yield of cool season grasses and clover species and cultivars under Michigan growing conditions.

Cool season grasses

Perennial ryegrass (*Lolium perenne* L.) is one of the most common cool season grasses grown in binary mixtures. It is considered a temperate perennial grass and is grown throughout the world including North and South America, Europe, New Zealand and Australia. It grows well in early spring and fall. However, during excessive high temperatures in summer months, it becomes dormant even with irrigation or abundant summer rainfall (Leep, 2004). Perennial ryegrass is suitable for pasture due to its high forage quality (Balasko et al., 1995). It can tolerate close frequent grazing, which makes it useful in an intensive grazing system (Jung et al., 1996). Perennial ryegrass has a higher IVDMD than other temperate perennial grass species (Pysher and Fales, 1992).

Orchard grass (*Dactylis glomerata* L.) is a major grass species commonly recommended for pastures in the Northeastern and North Central United States (Van Santen and Sleper, 1996) due to its greater drought tolerance and winter hardiness as compared to perennial ryegrass (Christie and McElroy, 1995). It is also well suited for mixtures with one or more legume species because of its fast growth and tillering ability (Hoveland, 1992). Tillering occurs almost continuously throughout the growing season with large number of tillers within a clump (the elongated stem). These remain vegetative, retaining the growth point close to the ground, which produce only leaves. Constant tiller production results in a rapid recovery after grazing (Christie and Mcelroy, 1995). Thus, orchard grass can persist under frequent grazing or clipping more than other grass species such as ryegrass, timothy (*Phleum pretense* L.), or tall fescue (Davies, 1988).

Festulolium (*Festulolium braunii, K.A.)* is derived from crosses between meadow fescue (*Festuca pratensis* Huds) or tall fescue (*Festuca arundinacea* Schreb.), and either Italian ryegrass (*Lolium multiflorum* Lam.) or perennial ryegrass. This cross combines the palatability of perennial or Italian ryegrass with the persistence of meadow or tall fescue. However, festulolium is less palatable and forms a more open sward as compared to perennial ryegrass (Wit, 1959). Festulolium combines the disease resistance and winter hardiness of meadow fescue with the high crude protein (CP) and good season-long productivity of Italian ryegrass. Winter hardiness of some festulolium varieties may approach that of perennial ryegrass (Ohio State University Extension bulletin).

Tall fescue (*Festuca arundinacea* Schreb.) is used widely for forage, turf, and conservation purposes (Collins and Hannaway, 2003). It is the predominant cool-season pasture grass in many southern states. It is tolerant of continuous close grazing and superior to many other cool-season grasses in livestock-carrying capacity. Tall fescue may be used as a hay crop, but needs to be harvested as the first seed heads begin to appear. Tall fescue is also grown as a turf grass species and its use has increased since the introduction of turf-type cultivars (Funk et al., 1981). Some tall fescue cultivars are infected with a fungal endophyte, *Neotyphodium coenophialum*, Margan-Jones & Gams, which produces alkaloids that are toxic to grazing animals (Ju et al., 2006) resulting in reduction of feed intake, weight gain, and milk production . However, non-endophyte and nontoxic endophyte-infected cultivars have been developed for livestock pastures. The nontoxic infected plants have shown improved tolerance to some environmental stresses such a drought and some insects (Collins and Hannaway, 2003).

Cool season clovers

Red clover (*Trifolium pratense* L.) is grown as a monoculture or mixed with grasses. Red clover may be grouped into three types: early-flowering, late-flowering and wild red clover. Most red clover growing in the US is the early-flowering type (Taylor and Smith, 1995). It is a very important legume hay crop in the Northeastern US. The relative performance of red clover cultivars may be different when monoculture and binary mixtures are compared. It is considered a short-lived perennial legume (Nelson and McGraw, 2003). Red clover is most often grown with a companion grass for hay, silage, or grazing. However, little information is available on the performance of red clover cultivars grown with grass species in rotationally grazed pastures. The performance of red clover cultivars is influenced by location, cutting system, and the presence or absence of grass.

White clover (*Trifolium repens* L.) (Common and ladino types) is one of the most important legumes used for grazing in the US. White clover is classified in three general cultivars groups: small, intermediate and large. The small cultivars are usually native and are referred to as wild white clover. Most white clover marketed in the US belongs to the intermediate type. Large cultivars referred to as Ladino, were introduced from Italy

(Henning and Wheaton, 1993). White clover should be grown in binary mixtures with grasses to prevent bloat in livestock. Under adequate soil moisture, white clover can grow in every state of the US and province in Canada (Pederson, 1995). White clover is very palatable and is high in forage quality (higher CP and lower fiber) and tolerates continuous, heavy grazing pressures (Spitaleri et al., 2003).

Kura clover (*Trifolium ambiguim* Bieb.) is a long-lived, perennial, rhizomatous legume (Bryant,1974; Taylor and Smith,1998) that tolerates frequent defoliation in monoculture (Peterson et al., 1994) or binary mixtures with grass (Kim,1996). It can be used for hay or pasture production (Sheaffer and Marten, 1991; Sheaffer et al., 1992). Kura clover is very compatible with grasses and suitable for mechanical harvest system (Kim, 1996). Kura clover grows slowly during the establishment year (Speer and Allinson, 1985). Slow establishment have limited widespread use of kura clover. This is related in part to the kura clover devoting most of its fixed C to roots and rhizomes during establishment and its slow nodulation, which limits N_2 fixation in the seeding year (Peterson et al., 1994; Seguin et al., 2001). However, with its rhizomatous root system, it is able to survive harsh environmental condition at northern latitudes (Pederson, 1995). DM yield of kura clover is lower than alfalfa but similar to red clover and birdsfoot trefoil (*Lotus corniculatus* L.) (Sheaffer and Marten, 1991). However, kura has higher forage quality (higher CP, lower acid detergent fiber [ADF] and neutral detergent fiber [NDF]) than other legumes (Allinson et al., 1985; Sheaffer and Marten, 1991). Kura clover has a higher risk of causing bloating in grazing animals when compared to white clover or alfalfa (Sheaffer et al., 1992). Because of this, it should be grown in combination with perennial grass to reduce incidence of bloat (Mouriño et al., 2003).

Grass and legume compatibility in binary mixtures is an important aspect of pasture systems. There are several factors that influence the ecology of grass-legume binary mixtures and many researchers have reviewed this subject. Grass and legumes compete for water and soil minerals when grown in mixtures (Jones, et al., 1988). However, competing for irradiance is often considered to be most critical among all the competition factors (Donald, 1961). Palatability of species can have a great impact on binary mixture composition. Animals tend to graze the most palatable species when multiple species are offered (Chapter 2). In addition, grass and legumes have different growth habits, regrowth, and physiological growth requirements, which make management more difficult for the binary mixtures compared to monocultures of the same species (Smith et al., 1986).

Forage grass species vary in their ability to persist with legumes in binary mixtures (Camlin, 1981). For instance, orchardgrass is more competitive in binary mixtures with alfalfa than smooth bromegrasses (*Bromus inermis*.Leyss) (Schmidt and Tenpas, 1965; Smith et al., 1973). This criterion is important when evaluating grass-legume associations (Zannone et al., 1986). Unlike alfalfa and red clover, white clover can tolerate both continuous and rotational grazing due to the proliferation of stolon segments that can grow and spread by forming new plants (Pederson, 1995). Under both rotational and continuous grazing, it is generally expected that DM yield of pastures will decrease to a certain extent during grazing season. Many factors can cause yield reduction. For example, changing botanical composition with a loss of legume can result in nitrogen deficiency. Soil structure and compaction can cause poor gaseous exchange at the root resulting in stand declines especially with legumes. In addition, soil nitrogen and potassium deficiencies as well as disease can also lead to DM yield reduction (Camlin and Stewart, 1976). It has also been reported that grass-legume balance is very susceptible to changes in respect to environmental conditions (Camlin, 1981). Therefore, for accurate assessment of grass-legume binary mixtures persistence, it is important for this type of research to be conducted under different soil types and environmental conditions.

Environmental conditions can regulate grass-legume interaction (Snaydon 1987). Low temperatures during the winter are an important factor in determining geographical distribution and persistence of forage species (Lorenzetti et al., 1971; Shimada et al., 1993). Winter injury occurs in most perennial grass and legume species and is related to the length and intensity of the cold weather conditions coupled with the effects of snow cover, soil heaving, and ice encasement. Bèlanger et al. (2002) reported that severe winters in eastern Canada caused recurrent damage to perennial forage crops. For a crop such as alfalfa, winter injury often occurs due to repeated freezing and thawing causing death of plants. Non-winter hardy species such as rye grass (*Lolium* sp L.) and festuloilum (some cultivars) have limited use in binary mixtures because of their poor cold persistence (Elissa et al., 1995). However, snow cover has a great impact on increasing forage survival. Leep et al. (2001) in Michigan concluded that 10 cm of snow cover is sufficient to protect alfalfa from winter injury. The ability to survive freezing temperatures depends upon adequately managing soil fertility, especially N and K, which can increase cold resistance (Nelson and Volenec, 1995). Winter survival of Coastal bermudagrass (*Cynodon dactylon*) decreased with increasing levels of applied nitrogen (N) fertilizer (Adams and Twersky, 1960). Genetic background may also influence the severity of winter injury (McKenzie et al., 1988). Past research indicates that tetraploid perennial ryegrass cultivars had significantly higher competitive abilities during summer and lower cold tolerances in winter than the diploid cultivars (Dvorak and Fowler, 1978; Sugiyama, 1998). Although the lower cold tolerance of tetraploid cultivars may lead to low persistence in pasture in northern latitudes, their lower survival rate in a cold winter is somewhat counterbalanced by high competitiveness during the summer (Sugiyama, 1998). Tetraploid cultivars of other forage species such as red clover and meadow fescue have been shown to have lower cold tolerance than diploids (Tyler et al., 1978).

In recent years, both the grass and legume seed industry have begun marketing new species and cultivars of perennial grasses and clovers to Michigan growers. For example, orchardgrass cultivars have been introduced with value-added traits such as increased tillering, later maturity, and greater winter hardness. Tall fescue has been recently offered endophyte free with smoother leaf blades. Most seed companies have information on how monocultures of their cultivars perform but they do not provide information on how the species perform as binary mixtures in long-term pastures. In addition, the ability of new cultivars to survive the severe winter conditions could be an issue, especially when grown in northern latitudes (44^{o}-46^{o} N). No previous research has been done to evaluate multiple grass and clover cultivars in binary mixtures at different latitudes in Michigan. Therefore, more research is needed to determine how these cultivars perform under Michigan growing conditions at latitudes 44^{o}-46^{o}.

In this study, several improved cool season grass and clover species and cultivars established in monoculture and binary mixtures were evaluated to determine their suitability for Michigan climatic conditions. Several aspects need to be considered for evaluating grass and clover species, including dry matter yield, winter injury, pasture ground cover, and clover composition in the mixtures.

MATERIALS AND METHODS

Experiment Establishment and Maintenance:

Monocultures and binary mixtures experiments were established in 2001 to be evaluated over a 5-year period (2002-2006) at three Michigan State University (MSU) experimental stations: (i) Kellogg Biological Research Station at Hickory Corners (KBS) (42° 24' N, 85° 24' W); (ii) The Beef Cattle and Forage Research Station at Lake City (LC) (44°19' N, 85°12' W) and (iii) Upper Peninsula Experiment Station at Chatham (UPES) (46°33' N, 86°55' W). The locations map is illustrated in Appendix Fig 1A. This research focuses on the period of 2004-2006.

The study area was separated into three portions: grass only, legume only (clover), and grass-clover binary mixtures (Appendix Fig. 2A). Each portion was a RCBD with three replications. For the grass-clover binary mixtures, each replication consisted of 67 entries of different combinations of grass and clover cultivars seeded in 1.8 by 5 m plots. Grass and clover monocultures experiments consisted of 16 and 8 grass and clover entries, respectively with plots of the same size as these of the binary mixtures portion.

Eight clover cultivars were established in this study included three red clover (VNS*, Star Fire, and Start), four white clover (KopuII, Ladino, Alice, and Jumbo), and one kura

* Variety Not Stated

clover (Endura). The 16 grass cultivars were included two festuloliums (Duo and Hykor), four orchardgrass (Amba, Niva, Sparta and Tekapo), seven perennial ryegrass (Aries, Maverick Gold, Quartet, Tonga, Barfort, Mara and Calibra) and three endophyte free tall fescues cultivars (Bronson, K5666V, and Barolex). Appendix Table 1A lists information on soil type, soil pH, harvest schedule, and grass and clover species and cultivars.

Soil was analyzed at the MSU soil-testing laboratory; fertilizers and lime were applied as recommended by soil test results. All tested soil parameters were at or above optimal levels (average: P: 60 ppm, K: 139 ppm, and pH: 6.5). In each location, nitrogen (N) was applied to the grass only plots following each cutting in the form of ammonium sulfate at a rate of 45.4 kg ha^{-1} in four applications through the growing season. However, since nitrogen deficiency appeared during 2005 in some grass species due to the poor clover stands in the binary mixture treatments, nitrogen was applied in a single application of (45.4 kg ha^{-1}) to the binary mixture treatments in all locations. Weeds were controlled with an herbicide in the grass monoculture treatments when necessary.

Weather record:

For each experimental location, total monthly precipitation (mm), average daily maximum and minimum air temperatures (^{o}C) during the months of growing season and the total monthly snowfall (cm) during winter months were recorded.

Data Collection and Sampling Procedure:

Three to four grazing (cutting) events were obtained from each location with 30 to 35 days interval between each event depending on weather conditions. The following data were collected:

Qualitative Measurements (Visual Estimates):

In all locations, qualitative measurements were recorded during 2004-2006. However, the clover monoculture portion was excluded in 2006 due to poor stand and weed contamination. Data on winter injury, percent living ground cover, and percent clover in the binary mixtures were obtained by visual rating estimates. The rating estimate was based on the average of the ratings generated by two trained people in each plot. The ratings were based on a 1 to 5 scale (Brummer and Moore, 2000). The scale system used for winter injury data collection is inversed from the one used either for ground cover or percentage clover. For examples, score 1 in winter injury is considered good (survive) and score 5 is poor (highly injured), while for ground cover and percentage clover data collection, score 1 is considered poor (low cover) and score 5 is good (highly covered).

Winter Injury: Winter injury ratings were taken during the early season growth (usually end of April to early May) for both monoculture and binary mixture treatments at all locations. A general percentage score of plots affected with winter injury was placed on the following scale: **1**= less than 20% of the vegetation killed; **2**= 20-40%; **3** = 40-60%; 4 = 60-80% killed; and **5** = greater than 80% of the vegetation was killed.

The cultivars that have low winter injury ranking over time are those that have high cold resistance.

Vegetative ground cover: Ground cover was recorded in the spring when the grasses were actively growing— two weeks after winter injury ratings were taken— and again in the fall before the dormancy period (at the end of October). Ground cover was visually scored by assessing the amount of vegetation cover based on the following scale: **1** = greater than 80% of the vegetation missing; **2** = 60-80%; **3** = 40-60%; **4** = 40 –20% and; **5** = less than 20% of the vegetation missing. The cultivars that have low ground cover ranking over time are those that have less persistence.

Percentage clover cover: Clover stands were visually rated for the percentage of clover versus grass in each plot of the binary mixture treatments. The ratings were conducted in the spring (early May) and fall (at the end of October) according to the following scale: **1**= less than 20% of the clover stand appears in the mixture; **2** = 20- 40%; **3** = 40-60%; **4** = 60-80%; and **5** = 80% or greater of the binary mixture was covered with the clover stand. The clover cultivars that have low cover ranking over time are those that are less compatible with grasses.

Quantitative Measurements:

Dry matter yield: Forage biomass of monoculture and binary mixture treatments were collected from all locations. When the average plant height was 20 cm, samples were hand clipped at 8 cm height within a 0.25 m^2 quadrate in each plot and dried at 60 °C for 48 hr. Dry samples were then weighted for DM yield.

Statistical analyses: All the statistical computations were performed using SAS (SAS Institute, 2000). Analysis of variance (ANOVA) was performed separately for binary mixtures, grass monoculture and clover monoculture using PROC GLM. The data from all years and locations were analyzed together. The interactions between years, locations, and cultivars (treatments) were examined and when found significant, mean separations were conducted using Fischer's LSD at $p<0.05$. Normality assumption was checked using stem and leaf and normal probability plots in PROC UNIVARIATE. Homogeneity of variance among years and locations was examined and found acceptable. Coefficient of variation (CV) was listed to measure the precision of the experiment. Winter injury was prevalent at LC in the spring of 2005 due to ice-sheeting in the lower portion of the experimental area, which resulted in serious damage in some plots. The affected plots were eliminated from the analyses and reported as missing data.

RESULTS AND DISCUSSION

I. Climatic Observations

Precipitation: In general, there was a high amount of precipitation recorded in 2004, especially during the months of May and August at the KBS and UPES locations. At LC, there was a higher amount of precipitation during the months of April and May and lower during the months of June, July, August and September. However, in 2005, lower precipitation was observed during most of the growing season at three locations with some higher amounts recorded in the month of July at KBS, August and September at LC and July and September at the UPES (Appendix Table 2A).

Average maximum and minimum daily air temperatures: At both KBS and LC locations, the maximum air temperature during 2004 was above the 30-year average only in April and September and in UPES, the maximum air temperature of 2004 was below the 30-year average throughout the growing season with the exception of September (Appendix Table 3A).

In 2005 growing season, the average maximum air temperatures at KBS, LC and UPES were above the 30-year average during the entire growing season except for the month of May (Appendix Table 3A).

The lack of precipitation along with above normal temperature in 2005 may have affected the DM yield for all cutting events. Average minimum air temperature (^{o}C) and snowfall (cm) data for the months of January through April of 2003-2006 is listed in Appendix Table 4A.

II. Persistence of Grass and Clover Cultivars Grown in Monoculture and Binary Mixtures

Persistence results include data obtained from winter injury and ground cover for each monoculture grass and clover cultivar and binary mixtures in all locations over 2004-2006.

Significant years X location X treatment interactions were present in winter injury, spring and fall ground cover for grass-clover binary mixtures (Appendix Table 6A). In addition, significant interactions were also present in grass monoculture treatment (varieties) for winter injury, spring and fall ground cover (Appendix Table 7 A), and similarly for clover monoculture treatment (varieties) (Appendix Table 8 A). We attribute these interactions primarily to weather conditions varying from locations to location and from year to year.

Grass-clover binary mixtures had less winter injury compared to monocultures of grass and clover (Fig1). Differences in winter injury score within cultivars and among locations were observed. Greater winter injury impact on grass cultivars occurred at the UPES as it is farther north than KBS and LC (Appendix Table 4 A). In 2006, the grass and clover species at LC were affected by ice-sheeting, resulting in some native grass such as Kentucky blue grass (*Poa pratensis*) to dominate the plots.

Analyses of winter injury of the grass monoculture showed there were significant differences ($p<0.05$) among the monoculture grass cultivars at KBS in all years (Table1). Grass cultivars displayed severe winter injury at the UPES location in the spring of 2006. Even winter hardy species like orchardgrass and tall fescue exhibited significant winter injury (Table1). Nitrogen applied the previous fall may have accentuated the winter injury. Adams and Twersky (1960) concluded that winter survival of coastal bermudagrass (*Cynodon dactylon*) decreased with increasing levels of applied nitrogen (N) fertilizer.

Cultivars that expressed severe winter injury (score 5) displayed less ground cover in their plots. Ground cover data of grass monoculture, grass-clover binary mixtures and clover monoculture are presented in Tables 3, 4 and 6, respectively. Winter injury and ground cover results will be discussed in more detail for each individual grass species.

Persistence of Grass Species

Festuloliums: There was no apparent winter injury with the two monoculture festulolium cultivars (Duo and Hykor) at the KBS. There was significant winter injury in Duo in 2006 (Table1). Duo resulted in significantly more winter injury ($p<0.05$) than Hykor, at the UPES site (4.3 and 5 in 2005 and 2006, respectively).

Analysis of festulolium-clover binary mixtures winter injury showed that significant winter injury occurred at the UPES during 2004-2006 compared to the same years at KBS and LC (Table 2). These data showed that persistence of Duo did not improve when associated with clover in binary mixtures at the UPES, while at KBS and LC, Duo showed improved winter survival when grown with clover in binary mixtures.

Analyses of ground cover of the monoculture festulolium cultivars (Duo and Hykor) showed both had excellent ground cover ratings (score 5) at KBS and LC.

However, ground cover was different between Hykor and Duo at the UPES with Duo resulting in significantly ($p<0.05$) less ground cover than Hykor during 2005 (Table3).

In conclusion, the results of winter injury and ground cover from the two festuloliums cultivars (Duo and Hykor) established in monoculture and binary mixtures showed Duo was less persistent and more vulnerable to winter injury when grown in the Upper Peninsula of Michigan. Duo has a ryegrass background (a hybrid resulting from a cross of meadow fescue and a tetraploid perennial ryegrass (AMPAC seed company web site). Ryegrass parents generally reduce winter survival of festuloliums hybrids and their derivatives (Casler et al., 2002), which helps explain the lower winter hardness of Duo. The lack of the adaptation of Duo in the upper peninsula of Michigan limits its use in pasture systems although it was preferred more by animals than Hykor (Chapter 2).

Orchardgrass: Winter injury data analyses showed that orchardgrass monoculture cultivars (Tekapo, Amba, Niva, and Sparta) had good winter survival at KBS and LC (Average score 1.3) (Table 1), which was similar to results reported by Christie and McElroy (1995). No significant differences in winter injury appeared at KBS and LC experimental locations, at the UPES, however, significant differences in winter injury between the four orchardgrass cultivars occurred during 2004-2006. Tekapo and Sparta demonstrated significant winter injury ($p<0.05$) at UPES (scores 4 and 5 respectively) when compared to the cultivars Amba and Neva.

Winter injury analyses of orchardgrass-clover binary mixtures showed a significant difference in winter injury at KBS with one treatment of Amba-red clover (VNS) in 2004 (Table 2). Higher winter injury was observed in Tekapo-clover binary mixtures at LC during 2006. The highest winter injury occurred in the Tekapo-Star Fire red clover binary mixture (score 3.7) (Table 2). Tekapo resulted in significantly higher winter injury when grown with all clover cultivars particularly with white clover Kopu II and Ladino compared to all other orchardgrass cultivars in 2006 at the UPES (Table2). These results indicate that Tekapo had significantly lower winter hardiness than all other orchardgrass cultivars even when grown with clover in binary mixtures.

Ground cover of the monoculture orchardgrass cultivars showed great vegetative cover at both KBS and LC (average score 4) compared to the same cultivars at the UPES in 2006 where scores ranged from 1 to 3 (Table 3). Tekapo resulted in significantly lower ground cover (score of 1 out of 5) than all other orchard grass cultivars due to winter injury.

Analysis of ground cover in orchardgrass-clover binary mixtures showed significantly lower ground cover for Tekapo when grown with white clover cultivars (Kopu II and Ladino) at UPES in 2006 (Table4). There were no significant differences in ground cover between orchardgrass cultivars grown in binary mixtures at the KBS, LC locations in 2004 and fall of 2005 at the UPES.

Orchardgrass is quite winter hardy when compared to perennial ryegrass (Christie and McElroy, 1995). In this study, four orchard grass cultivars grown in monoculture and binary mixtures exhibited good ground cover and persistence when grown at the KBS and LC experimental stations. At the UPES location, however, there were differences in persistence and ground cover among cultivars as a result of colder temperatures. Tekapo resulted in significantly lower persistence at the UPES site whether grown in monoculture or binary mixtures.

Perennial ryegrass: Perennial ryegrass cultivars were more susceptible to winter injury. However, analysis of perennial ryegrass monoculture showed there was no significant winter injury in the first two years of the study at KBS, but Maverick Gold had significantly higher winter injury in 2006 (Table1). At LC, there were significant differences between perennial rye grass cultivars in all years. Calibra showed significantly higher persistence in each year at LC compared to other cultivars. Calibra resulted in significantly higher winter hardiness and better persistence at the UPES location compared to other cultivars; however, all cultivars including Calibra were completely killed in 2006. Aries, Maverick Gold, and Quartet were all completely killed in 2005 with an injury rating of 5 while Calibra resulted in an injury rating of 3. The remaining cultivars were not significantly different in winter injury. The cultivars, Mara, Tonga, Calibra and Barfort had slightly less winter injury at LC and UPES in 2004.

Perennial ryegrass-clover binary mixtures resulted in less winter injury compared to the same cultivars grown in monoculture. Maverick Gold-red clover (VNS) binary mixtures at KBS had significantly greater winter injury than other entries (Table 2). Similarly, data from the LC site showed significantly higher winter injury ($p<0.05$) for the binary mixture of Aries-kura (Endura). All binary mixtures of Maverick Gold-clovers and Quartet-kura (Endura) resulted in significantly higher winter injury ($p<0.05$) at LC (Table 2). At the UPES, the perennial ryegrass-clover mixtures of Maverick Gold had significantly less persistence than all other treatments in 2004. Some treatments of

Calibra, Mara, and Tonga grown in binary mixtures resulted in significantly less winter injury compared to other binary mixtures in 2005, however, there was considerable winter injury of those cultivars in 2006 (Table 2).

Ground cover analysis of the perennial ryegrass cultivars showed better persistence of cultivars established at the KBS and LC sites compared to UPES site (Table 3). Aries had significantly higher in ground cover than Maverick Gold ($p<0.05$) at KBS in fall 2005 and spring 2006 (Table 3). At LC, lower ground cover observed for Maverick Gold, Aries and Mara in spring of 2005 and Calibra and Quartet resulted in significantly higher ground cover in spring of 2006 compared to all other perennial ryegrass cultivars (Table 3). At the UPES, significant lower ground cover was found in Aries, Maverick Gold and Quartet during spring 2005 and significantly lower for all cultivars in 2006 (Table 3).

Ground cover analysis of perennial ryegrass-clover binary mixtures presented in Table 4 show high ground cover at KBS and LC in 2004-2005 even with less winter-hardy cultivars. There was no significant difference found for ground cover at KBS. However, at LC and UPES, significant differences were found between cultivars in 2005. Analysis of the data from the UPES site showed good ground cover ratings in 2004, however, there were low ground cover ratings for all binary mixtures of perennial ryegrass cultivars in 2005 (Table 4). In spring, 2005, all perennial-clover binary mixtures were significantly lower in ground cover compared to binary mixtures of Calibra (Table 4). By fall 2005, all cultivars recovered from winter injury and were higher in ground cover. However, in the spring of 2006, all cultivars were again lower in ground cover with ratings of less than 50 % of the fall, 2005 ratings.

In summary, there was greater persistence when perennial ryegrass cultivars were grown in binary mixtures compared to the same cultivars grown in monoculture. Perennial ryegrass cultivars such as Maverick Gold, Aries, and Quartet had significant higher winter injury at LC and UPES, where colder winter temperatures prevail compared to the KBS location.

Tall fescue: Winter injury analyses of the three tall fescue monoculture cultivars (Bronson, K5666V and Barolex) showed Bronson to have significantly less winter injury than Barolex at LC in 2005. No significant differences were found between tall fescue cultivars at LC in 2006. However, in 2004, Bronson was significantly lower winter injury than K5666V and Barolex (Table 1). There was more winter injury at the UPES site for all cultivars in 2006 with scores ranging from 3 for Bronson and 5 for K5666V and Barolex. Bronson resulted in significantly less winter injury than K5666V and Barolex at the UPES in 2006 (Table 1). These data show Bronson has significantly higher winter hardiness than the other cultivars in this study. Bronson is well adapted to all latitudes in Michigan.

Winter injury analyses of tall fescue-clover binary mixtures from at locations showed little winter injury (Table 2). However, K5666V resulted in significantly higher winter injury than Bronson when grown with 3 of the 5 clover species and cultivars in 2005 at LC and UPES.

Ground cover of all tall fescue monoculture showed excellent cover across all locations (Table 3). However, at LC, Barolex was significantly higher in ground cover than K5666V and Bronson in spring 2005. K5666V was significantly lower in ground cover compared to Bronson at UPES in 2005 while both K5666V and Barolex were significantly lower in ground cover than Bronson in the spring of 2006 at the UPES (Table 3).

Ground cover analysis of tall fescue-clover binary mixtures is presented in Table 4. There was higher ground cover for all years and locations (score 4 to 5) with binary mixtures compared to monoculture of tall fescue. Bronson tall fescue grown in binary mixtures of Ladino white clover and VNS red clover was significantly higher in ground cover in 2005 at the UPES compared to other binary mixtures and tall fescue cultivars. There were no differences in ground cover between cultivars and species of clovers in any other treatments and years. These data indicate better ground cover when tall fescue is grown with clovers compared to tall fescue grown in a monoculture.

In conclusion, the endophyte free tall fescue cultivars used in this study demonstrated an excellent persistence whether grown in monoculture or binary mixtures at KBS, LC for all years and 2004-2005 at UPES. Binary mixtures of tall fescue –clover showed greater winter hardiness and ground cover at all locations, which is likely due to better distribution of available nitrogen throughout the growing season for tall fescue growing in association with clovers.

Persistence of Clover Species

Kura clover had significantly less winter injury than some red and white clovers cultivars at KBS. No data were recorded at LC during 2005-2006 due to poor stands with the exception of kura clover (Endura), which did not have winter injury (Table 5). However, there was little winter injury in all cultivars at LC during 2004. Star Fire resulted in significantly higher winter injury in 2004 compared to other cultivars. At UPES, kura clover had the greatest winter injury (score 3.8) among all clover cultivars (Table 5). However, this may be an anomaly as kura clover has been shown to have a high winter hardness. It is possible these results were attributed to poor establishment. Overall, there was less winter injury in the clover monocultures compared to the perennial grass monoculture.

Ground cover of all clover cultivars was high (scored 3.5 to 4) at KBS in all years Table 6). There were significant differences in ground cover between species or cultivars at KBS. At LC site, there were significant differences between treatment means in ground cover in fall of 2004. The ground cover of kura clover at LC in the spring of 2005 resulted in a high score (3.2) (Table 6). In contrast, kura clover had low ground cover in fall of 2004 at LC, which is likely related to an initial poor stand of kura clover at seeding time due to slower establishment (Cuomo et al., 2003). At the UPES, data analyses showed significantly higher ground cover scores for all white clover cultivars (Kopu II, Alice, Jumbo and Ladino), which was likely due to excellent stolon production resulting in new plants forming. However, diseases such as root rots eliminated red clover stands (Taylor and Smith, 1995), which helped explain the low ground cover scored at the UPES (scored of 1.8) (Table 6) in spite of their persistence to cold winter.

At LC, higher ground cover for all the eight cultivars was found during 2004. Red and white clovers did not persist and resulted in no ground cover in the last two years (2005-2006).

III. Grass-Clover Composition in Binary Mixtures

Significant year X location X treatment (binary mixtures) interactions were present for spring and fall clover content (Appendix Table 6A). These interactions resulted primarily from changes in mixture compositions that occurred from year to year due to low persistence of some clovers. Statistical analysis was preformed for the entire 67 binary mixture treatments in this study. However, results were reported separately by grass species. Results presented in Tables 7, 8, 9, and 10 are the mean values of percentage clover grown with festuloliums, orchardgrass, perennial ryegrass and tall fescue respectively.

Clover Composition in Festulolium -Clover Binary Mixtures: There was a significant difference ($p<0.05$) in clover composition between the clover cultivars grown with Duo (Table 7). The clover species and cultivars grown in association with festulolium cultivars resulted in different responses from year to year and location to location. In the fall of 2004 (three years after establishment), kura clover cultivar (Endura) composition surpassed 80% (scored 5) when grown with Duo at KBS. However, kura clover composition growing with festulolium (Duo) was significantly less at both LC and UPES locations (scored 2.4 and 1.9 respectively) (Table 7). Analysis of the data showed that red clover cultivars (Star Fire, VNS) at KBS had a high clover composition (scored 4 to 5) when associated with the two festulolium cultivars during the first three years of production followed by a progressive decline in their composition (scored 1 to 1.7) (Table 7).

The LC site showed a high clover composition in the fall of 2005, however, much of this was from volunteer white clover, which filled in after the red clover plants died. As mentioned previously, red clover is susceptible to root rot diseases, which cause a decline in their stands (Taylor and Smith, 1995). There was low clover composition of white clover cultivars (Jumbo and Ladino) in all locations during spring 2005 and spring 2006 when associated with Hykor (scored 1 to 1.7) (Table 7). Hykor was more dominant in the binary mixture, which contributed to the lower clover botanical composition.

In conclusion, Endura kura clover botanical composition increased each year when grown with festulolium (Duo) binary mixtures at the KBS location. However, this was not the case at LC or UPES locations where kura clover stands were not as competitive as kura established at KBS. A possible reason may be related to the poor establishment at LC and UPES, which resulted in lower stands over time. It may also be attributed to the highly competitive festuloliums, which grow more vigorously at northern locations.

Clover Composition in Orchardgrass-Clover Binary Mixtures: There was a significant difference ($p<0.05$) in clover botanical composition between orchardgrass-clover binary mixture treatments (Table 8). Previous research has suggested that orchardgrass is well suited for mixtures with one or more legume species (Hoveland, 1992). Orchardgrass is also characterized by the extensive tillering that occurs throughout the growing season, enabling the species to have quick re-growth following harvest or grazing. This vigorous growth makes this species highly competitive in binary mixtures (Casler, 1988).

Red and white clover cultivars in binary mixtures with the orchardgrass cultivars were lower in composition during the final two years of the study at KBS and UPES (scored 1 to 1.7) (Table 8). This may be due to less persistence in red clover caused by root rots or winter injury in white clover. At LC, however, there was moderate clover composition in the mixtures of red (VNS) and white clover (Ladino) stands when grown with Niva and red clover (Star Fire) when growing with Tekapo (Table 8). In contrast, kura clover (Endura) resulted in significantly higher composition at the KBS location when grown with Tekapo, which persisted throughout all years in this study (scored 4.3) (Table 8). These results are supported by the observations of Hoveland (1992) and Casler (1988), which concluded that orchardgrass is more competitive in binary mixtures with clovers. The quick re-growth following harvest enables orchardgrass cultivars to be more dominant in the binary mixtures. In addition, with adequate moisture, kura clover is considered an excellent choice for Southern Michigan with orchardgrass in binary mixtures. However, slow establishment of kura clover made it less competitive with orchardgrass at LC and UPES. Perhaps if kura was seeded by itself and allowed to establish first followed by seeding the perennial grass, it would result in good stands.

Clover Composition in Perennial Ryegrass-Clover Binary Mixtures: Perennial ryegrass- clover binary mixtures were significantly different ($p<0.05$) in clover composition between the seven perennial ryegrass cultivars used in this study at KBS in the fall 2005 and spring 2006 (Table 9). Significant differences in the clover composition of perennial ryegrass- clover was found in LC, spring and fall, of 2004 at UPES. There were significant differences in clover composition between perennial-clover binary mixtures for all years. At KBS, kura clover (Endura) had the greatest clover composition among perennial ryegrass cultivars over all years (average score 4). These results were different at the other locations.

At LC, the clover composition during spring 2005 of white clover (Kopu II and Ladino) was higher when grown with Tonga. Red clover (Star Fire) composition was greater than other clovers when grown with Quartet. Clover composition of white clover (Ladino) was greater than other clovers when grown with Barfort, Tonga, Maverick Gold and Quartet at the UPES site. Alice and Kopu II white clover clover composition was higher when grown with Mara and Maverick Gold perennial ryegrass respectively (Table 9). All red clovers growing with perennial ryegrass cultivars at KBS and UPES persisted well until the fall of 2005 but nearly disappeared by the spring of 2006 with average percentage cover rating of 1.5 (Table 9). This was likely due to root rots, which red clovers are susceptible to. On the other hand, the increase of clover composition in red clover observed in LC binary mixtures was likely related to the appearance of volunteer native legume from the soil seed bank. White clover cultivars persisted similarly to red clovers at KBS (Ladino, Kopu II, Alice and Jumbo) with good clover cover until the spring of 2006.

In conclusion, kura clover's persistence was excellent with all perennial ryegrass cultivars at the KBS location and though to a lesser extent at the LC site. A study conducted by Cuomo et al. (2003) in Minnesota showed that kura clover was able to compete, persist, and spread under intermittent grazing and has the potential to be an important and persistent component of cool-season grass pasture in north-central USA. However, white clovers, in particular, Kopu II and Ladino were higher in clover composition (scored 3 to 4) at the UPES site compared to red clover and kura clover cultivars. The initial stand of kura clover at the UPES site was not as good as the other two locations, which may have contributed to the lower botanical composition with perennial grasses at this site.

Clover Composition in Tall Fescue-Clover Binary Mixtures: Tall fescue cultivars (Barolex, K5666V and Bronson) were more competitive with all clover species at all locations (average scores from 1.3 to 3) (Table 10). Results showed kura clover composition was significantly greater than all other clovers when grown in binary mixtures with K5666V cultivar at the KBS location (average score 3.9) (Table 10).

Tall fescue was more aggressive when grown with all clover species compared to some cultivars of festulolium, orchardgrass, and perennial ryegrass. Tall fescue resulted in good stand persistence at all locations. This species should be considered when developing new pastures. However, according to this study, white and red clover cultivars may need to be reseeded more often when associated with tall fescue with the exception of kura clover, which showed better compatibility than other species when grown with tall fescue (K5666V) at KBS (Table 10).

In conclusion, binary mixtures behaved differently depending on the grass and clover growing in the mixtures. There was variation in clover composition among grass cultivars and years. For example, some grass cultivars that had low clover composition in LC in the first two years showed excellent clover composition in the fall of 2005 such as festuloliums (Duo)-red clover (Star Fire) (Table 7), orchardgrass (Niva)-red clover (VNS) (Table 8), and most perennial ryegrass cultivars (Table 9). This was likely due to winter injury in grass cultivars resulting in little or no grass competition and higher composition of clover in these binary mixtures.

IV. Dry Matter Yield of Grass And Clover Cultivars Grown in Monoculture And Binary Mixtures.

Seasonal differences in DM production of species and cultivars occurred across grazing events and locations. There was also an overall decline in DM production in time across species. Significant year X location X cut X treatment (binary mixtures) interactions were observed in DM yield (Appendix Table 9 A). In addition, significant year X location X cut X treatment (varieties) interactions in DM yield was found in clover monocultures (Appendix Table 11 A). These interactions are primarily due to changes in DM yield production resulting from different weather conditions, locations and variation among cultivars. DM yield (tons acre^{-1}) analyses were reported for each individual location, year, and cutting event. However, there was no interaction between grass monoculture treatments (Appendix Table 10 A). These data are presented by location, where Tables 11, 12 and 13 present the mean values of the grass monoculture yield at KBS, LC and UPES respectively; Tables 14, 15 and 16 give the mean yield values of the grass-clover binary mixtures at KBS, LC and UPES respectively and Tables 17, 18, and 19 provide the mean yield values of clover monoculture at KBS, LC and UPES locations respectively.

Dry Matter Yield of Grass Monocultures

Yield (tons acre $^{-1}$) analyses of grass monocultures are presented in Tables 11,12 , and 13. Cultivar and species varied in DM yield. DM yield for each individual grass type within a cutting event are presented in details as follows:

Festulolium: The monoculture Duo and Hykor were significantly different in DM yield. Hykor had significantly higher ($p<0.05$) DM yield than Duo in four of eight cutting events at KBS during 2004 and 2005 growing season (Table 11). LC dry matter yield analysis showed Hykor was significantly higher ($p<0.05$) than Duo in three of six cutting events during 2004-2005 (Table 12). Hykor was also significantly higher at two of eight grazing events during 2004-2005 at UPES (Table 13). Total two year DM yield of Hykor was significantly higher than Duo at all locations. Total DM yield production of Duo festulolium at the UPES site was greater than either KBS or LC (Tables. 11, 12 and 13), which indicates that Duo performs better in a cooler climate such as Northern Michigan.

Orchardgrass: There was a significant difference in DM yield between monoculture orchardgrass cultivars in one of eight cutting events at KBS (Table 11). There was a significant difference in DM yield in two cutting events between orchardgrass cultivars at LC and UPES (Table 12 and 13 respectively). Total two-year DM yield was numerically greater (tons acre $^{-1}$) in each monoculture orchardgrass cultivar at UPES compared to KBS and LC.

Perennial ryegrass: DM yield ranged from 2.37 tons acre $^{-1}$ for Maverick Gold to 5.92 tons acre $^{-1}$ for Mara at the UPES location (Table 13). Lower DM yield resulted from

Maverick Gold since it was severely affected by winter injury at the UPES location in 2005 (Table 13). Some of the Maverick Gold stands were recovered, but did not contribute significantly to yield. Statistically, there was a significant difference in DM yield between monoculture perennial ryegrass cultivars in three of eight cutting events at KBS (Table 11). At the LC site, a significant difference in DM yield occurred in two cutting events (Table 12). There was a significant difference in DM yield in six of seven cutting events at UPES (Table 13). DM yield of perennial ryegrass was excluded from the fourth cutting event (October) in 2004 at UPES due to lack of moisture that occurred in perennial ryegrass cultivars at the UPES (Appendix Table 2A).

Tall fescue: Monoculture tall fescue resulted in the highest DM yield among all grass species. DM yield ranged from 3.95 with K5666V at the KBS site (Table 11) to 6.88 tons acre $^{-1}$ with Barolex at the UPES (Table 13). High DM yield productivity (average of 1.41 tons acre $^{-1}$) for all grass cultivars was obtained at the second cutting event at UPES in 2004 (Table 13). There was a significant difference in DM yield between monoculture tall fescue cultivars in four of eight cutting events at KBS (Table 11). At LC, a significant difference in DM yield occurred in one of six cutting events (Table 12). At the UPES, there was a significant difference in DM yield in two cutting events during 2004-2005 (Table 13).

Dry Matter Yield of Grass-Clover Binary Mixtures

There were significant differences in DM yield of festulolium Hykor- clover binary mixtures compared with Duo-clover mixtures and results varied between locations. For example, Hykor-Ladino binary mixtures were significantly higher in DM yield compared to Duo-Ladino mixtures in four of eight cutting events at KBS (Table 14), while there was no significant difference between these mixtures at the LC site (Table 15). At the UPES there was a significant difference in two cutting events (Table 15). In addition, Hykor grown in binary mixture with white clover (Jumbo) and red clover (VNS) resulted in a significantly higher DM yield at KBS site (Table 14). There were significantly higher DM yield in Hykor binary mixtures at KBS (Table 14) and LC sites (Table 15) compared to Duo-binary mixtures. However, Duo- Star Fire red clover binary mixture was significantly higher in DM yield than Hykor-red clover (VNS) and white clover (Ladino) at the UPES site (Table 16).

Orchardgrass- clover binary mixtures were higher in DM yield at KBS (Tables 14) and LC (Tables 15) compared to the UPES site (Tables 16). Sparta grown with VNS red clover was significantly higher yielding than other orchardgrass-clover binary mixtures at the KBS location (Table14). Tekapo grown with VNS red clover had a significantly higher DM yield at the LC location (Table 15). Orchard grass (Sparta)-jumbo white clover binary mixtures were the highest in DM yield. Tekapo associated with Endura kura clover was the lowest yielding among the orchardgrsas-clover binary mixtures at the UPES location (Table 16). Even though orchardgrass is considered an excellent cool season grass for binary mixtures due to its fast re-growth and tillering ability (Hoveland, 1992), there are some factors, which might limit its success. For instance, winter injury of some orchardgrass-clover binary mixtures such as Tekpao-kura (Endura), and Tekapo growing with white clover (Kopu II and Ladino) were lower in DM yield at the UPES site (Table 16). Since orchardgrass is an aggressive grass, it may crowd out associated clovers growing in binary mixtures, thus causing reduction in clover content with resultant nitrogen deficiency from lack of fixed nitrogen from lower clover content.

Perennial ryegrass- clover binary mixtures were highly in DM yield compared to monoculture treatments of perennial ryegrass at all locations. In all locations, the total DM yield ranged from 2.05 tons acre^{-1} in Maverick Gold- kura (Endura) at UPES (Table 16) to 5.36 tons acre^{-1} in Calibra-red clover (VNS) at LC (Table 15).

The UPES site resulted in lower DM yield of perennial ryegrass-clover binary mixtures. Winter injury of non hardy perennial ryegrass cultivars was likely the reason for the lower yield at this location. The low yield of the Maverick Gold-Endura kura clover binary mixture in the UPES was likely related to a combination of winter injury to the Maverick Gold perennial ryegrass and poor establishment of Endura kura clover. Likewise, at the UPES the higher yield of Tonga-Star Fire red clover was likely due to greater winter hardiness of Tonga perennial ryegrass and more vigorous growth of Star Fire red clover (Table 16). Mara perennial ryegrass grown with VNS red clover had significantly higher DM yield than Aries-Endura kura clover, Quartet-Endura kura clover, and Quartet-Kopu II at the KBS location.

Tall fescue- clover binary mixtures ranged from 3.90 to 6.29 tons acre^{-1} in DM yield for K5666V- Koppu II and Bronson-Endura kura clover treatments, respectively at the KBS location (Table 14). DM yield of Barolex-Start was significantly higher than Barolex-Alice but not different than other Barolex-clover binary mixtures at KBS (Table 14).

In general, KBS, tall fescue cultivars grown in binary mixtures with clovers performed similarly to orchardgrass- clover cultivars in yield but were higher than both perennial ryegrass and festulolium- clover mixtures. For LC, DM yield of tall fescue-clover binary mixtures ranged from 3.96 to 5.33 tons acre^{-1} for K5666V-Star Fire red clover and Bronson-VNS red clover mixes (Table 15) and for the UPES, DM yield of tall fescue- clover binary mixtures ranged from 2.81 to 4. 90 tons acre^{-1} with Bronson-Endura kura clover and Barolex-Start red clover mixtures (Table 16).

The average total DM yield of tall fescue- clover binary mixtures was greatest at the KBS location followed by LC and UPES locations. Tall fescue-clover binary mixtures appear to be a good choice for all the locations in this study. These data indicate good yield stability for tall fescue-clover binary mixtures in Michigan However, as mentioned previously, it is important to select the proper cultivars of both tall fescue and clovers as each performed differently depending on location.

Dry Matter Yield of Clover Monocultures

Dry matter yield of clover monoculture at KBS is presented in Table 17, at LC in Table18 and at UPES in Table 19. There was a significant difference ($p<0.05$) in DM yield observed among clover species and cultivars within cutting events at KBS, UPES 2004-2005 and LC 2004. Endura kura clover resulted in significantly higher total DM yield than red clover (VNS) and white clover (Kopu II, Alice and Ladino) KBS (Table 17). At LC, DM yield ranged from 1.95 to 3.24 tons acre^{-1} at LC with Endura kura clover being also the highest in DM yield (Table 18). At UPES, no significant difference was found in total two year DM yield between clover species and cultivars (Table 19).

These data clearly showed Endura kura clover to be well adapted to southern Michigan where it had the highest yield of all clover cultivars in this study. Some cultivars of clover such as red clover (VNS) and white clover (Kopu II) had higher total one-year production at LC (Table 18) than the same cultivars produced in two years at the UPES site (Table 19).

CONCLUSIONS

Grass-clover binary mixtures resulted in increased DM yield and had higher resistance to winter injury compared to grasses and clovers in monoculture. However, this study demonstrated the importance of proper species and cultivar selection.

Grass and clover species in binary mixtures showed significant differences between species and cultivars. In the early part of this study, there was a balance in botanical composition between grass and clover content. However, this balance disappeared after two seasons with either the grass or clover soon becoming more or less dominant. Vigorous grass species such as tall fescue and orchardgrass tended to be more dominating in the mixtures resulting in decreased clover content. In contrast, some clovers persisted better than grasses. Environmental conditions are a factor that changes the grass-clover balance (Camlin, 1981). Thus, winter injury leading to a loss in vegetative ground cover of some grasses, especially perennial ryegrass provided an opportunity for the associated clover to dominate the mixtures. When clover disappeared from the binary mixtures, there was limited nitrogen (N) fixation, which resulted in nitrogen deficiency and subsequent loss of DM yield of the associated grass species. Therefore, when the clover content becomes low in the mixtures, nitrogen application is necessary to maintain high DM yield. This study demonstrated the importance of compatible clovers and grasses species in binary mixtures. Non-winter hardy cultivars of grass such as perennial ryegrass (Maverick Gold and Aries) used in this study showed greater persistence when they were grown in binary mixtures due to reduced winter injury.

Nitrogen fixation by clovers has a positive effect upon winter survival of perennial grasses. In binary mixtures, available nitrogen from N fixation of clover is provided to grasses in the critical spring and summer months in a steady supply. However, in the fall months, N fixation slows in clovers with subsequent less N available to grasses resulted in hardening of grasses and better winter hardness. Insulation from snow cover also increases grass survival in the winter (Leep et al., 2001). Growing grass with clover in binary mixtures can help to intercept snow resulting in less winter injury.

Binary mixtures of clover and grass resulted in a higher total DM yield and more uniform dry matter distribution through the season. These results are similar to those observed by Chamblee and Collins (1988).

Tall fescue resulted in excellent yield stability and broad adaptation to different environments compared to other grass species in this study, while perennial ryegrass yield was found to be unstable. However, there were significant differences in the performance of perennial ryegrasses cultivars due to differences winter hardiness.

Kura clover (Endura) provided the highest DM yield and exceptional persistence under grazing compared to other clover cultivars in this study, particularly at 42° N latitude zone. Thus, the influence of latitude on the performance of clover was observed in this study. Kura clover (Endura) demonstrated the highest persistence, which is similar to results found by Woodman et al., (1992). However, the establishment of kura is somewhat difficult, as it grows more slowly during the establishment year than other clover species (Speer and Allinson, 1985). For northern latitudes, kura clover dry matter yield was low due to the poor establishment, which indicates that improved establishment of this species need more attention for it to be successful in binary mixtures. Interseeding cool-season grass species into established kura clover provides an opportunity for kura to be well established prior to grass establishment.

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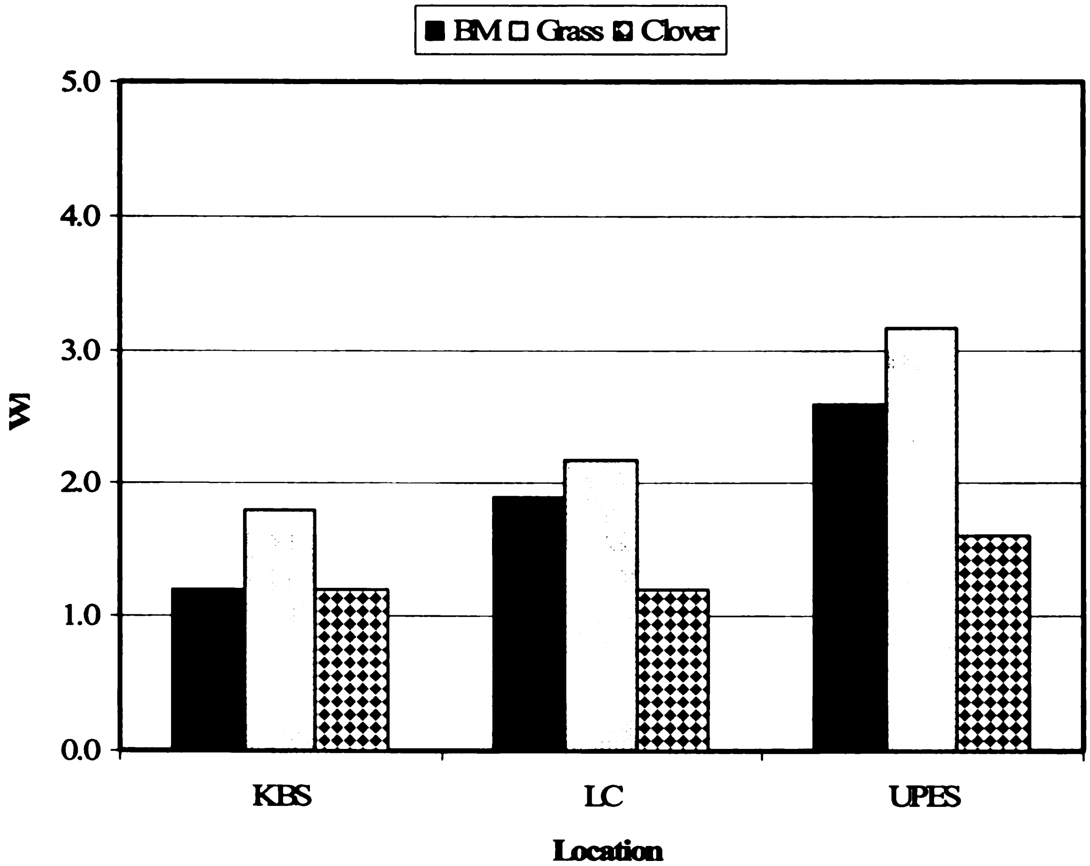

Fig.1. Comparing average winter injury (WI) at Kellogg Biological Research (KBS), Lake City (LC) and Upper Peninsula Experiment Station (UPES) for grass-clover binary mixtures (BM), grasses, and clovers grown in monocultures.

1= less than 20% of the vegetation was killed.
5= Greater than 80% of the vegetation was killed.

Table 1. The mean values of winter injury rating of 16 grass cultivars established in monoculture at Kellogg Biological Research Station (KBS), Lake City (LC) and Upper Peninsula Experiment Station (UPES) over a three-year period.

Grass	Grass	KBS				LC				UPES			
Species	**cultivars**	2004	2005	2006	3-yr ave	2004	2005	2006	**3-yr ave**	2004	2005	2006	**3-yr ave**
Fest	Duo	1.6	1.0	1.0	**1.2**	1.6	2	2.3	**2.0**	1.8	4.3	5.0	**3.7**
	Hykor	1.3	1.0	1.0	**1.1**	1.0	1.0	1.0	1.0	1.3	1.0	2.0	**1.4**
OR	Tekapo	1.5	1.0	1.0	**1.2**	1.0	2.7	1.0	**1.6**	1.3	3.0	5.0	**3.1**
	Amba	1.1	1.0	1.0	**1.0**	1.0	1.4	1.0	**1.1**	2.0	2.0	2.8	**2.3**
	Niva	1.0	1.0	1.0	**1.0**	1.1	1.5	1.0	**1.2**	1.0	1.6	3.3	**2.0**
	Sparta	1.0	1.0	1.0	**1.0**	1.6	1.4	1.0	**1.3**	1.0	1.3	4.0	**2.1**
PR	Aries	1.8	1.2	1.2	**1.4**	3.0	4.4	5.0	**4.1**	4.1	5.0	5.0	**4.7**
	Mvrck Gld	2.0	1.8	4.0	**2.6**	4.0	3.7	4.3	**4.0**	4.0	5.0	5.0	**4.7**
	Quartet	2.1	1.0	1.0	**1.4**	3.0	3.7	4.0	**3.6**	2.6	5.0	5.0	**4.2**
	Tonga	1.3	1.0	1.0	**1.1**	1.8	3.0	3.0	**2.6**	2.1	4.0	5.0	**3.7**
	Barfort	1.5	1.0	1.0	**1.2**	2.0	3.0	3.0	**2.7**	2.0	4.0	5.0	**3.7**
	Mara	1.5	1.0	1.0	**1.2**	2.6	4.4	3.3	**3.4**	2.0	4.0	5.0	**3.7**
	Calibra	1.3	1.0	1.0	**1.1**	2.0	2.7	1.3	**2.0**	1.8	3.3	5.0	**3.4**
TF	Bronson	1.0	1.0	1.0	**1.0**	1.0	1.0	1.0	1.0	1.0	1.3	3.5	**1.9**
	K5666V	1.5	1.0	1.0	**1.2**	1.1	1.4	1.0	**1.2**	1.5	2.8	5.0	**3.1**
	Barolex	1.6	1.0	1.0	**1.2**	2.0	2.5	1.3	**1.9**	2.0	2.3	5.0	**3.1**
	Mean	1.4	1.1	1.2		1.6	2.5	2.2		1.8	3.1	4.4	
	CV	27.2	9.9	6.0		35.9	33.0	23.9		34.8	15.4	5.9	
	LSD (0.05)	0.6	0.2	0.2		0.9	1.4	0.8		1.1	0.8	0.4	

1= less than 20% of the vegetation was killed.
5= Greater than 80% of the vegetation was killed.
Fest=Festulolium ; OR=Orchardgrass, PR=Perennial ryegrass and TF= Tall fescue.

Table 2. The mean values of winter injury rating of 67 grass-clover binary mixture treatments established at Kellogg Biological Research Station (KBS), Lake City (LC) and Upper Peninsula Experiment Station (UPES) over a three-year period (continued on the next page).

Grass	Clover	KBS				LC				UPES			
Cultivars	**Cultivars**	'04	'05	'06	**AVE**	'04	'05	'06	**AVE**	'04	'05	'06	**AVE**
Duo	Endura	1.6	1.0	1.0	**1.2**	1.5	1.3	2.3	**1.7**	2.0	4.0	4.7	**3.6**
Duo	Star Fire	1.6	1.0	1.0	**1.2**	1.0	2.8	1.7	**1.8**	2.1	4.3	4.7	**3.7**
Duo	VNS	1.6	1.0	1.0	**1.2**	1.6	1.0	2.7	**1.8**	1.8	3.7	4.5	**3.3**
Duo	Ladino	2.0	1.0	1.0	**1.3**	1.3	2.5	3.5	**2.4**	1.5	4.7	4.7	**3.6**
Duo	Kopu II	1.6	1.0	1.0	**1.2**	1.2	2.0	2.7	**2.0**	1.3	4.3	4.8	**3.5**
Hykor	VNS	1.0	1.0	1.0	**1.0**	1.3	1.0	1.0	**1.1**	1.0	1.3	1.0	**1.1**
Hykor	Jumbo	1.5	1.0	1.0	**1.2**	1.5	1.6	1.3	**1.5**	1.0	1.2	1.0	**1.1**
Hykor	Ladino	1.0	1.0	1.0	**1.0**	1.1	1.0	1.0	**1.0**	1.0	1.5	1.0	**1.2**
Amba	VNS	1.8	1.0	1.0	**1.3**	1.3	1.1	2.3	**1.6**	1.0	1.0	1.0	**1.0**
Amba	Jumbo	1.6	1.0	1.0	**1.2**	1.0	1.3	1.0	**1.1**	1.0	1.2	1.7	**1.3**
Amba	Ladino	1.0	1.0	1.0	**1.0**	1.1	1.0	1.3	**1.1**	1.1	1.0	1.3	**1.1**
Niva	VNS	1.1	1.0	1.0	**1.0**	1.1	1.1	2.0	**1.4**	1.0	1.0	1.0	**1.0**
Niva	Jumbo	1.1	1.0	1.0	**1.0**	1.0	1.5	2.3	**1.6**	1.0	1.0	1.7	**1.2**
Niva	Ladino	1.0	1.0	1.0	**1.0**	1.7	1.6	2.3	**1.9**	1.0	1.0	1.3	**1.1**
Sparta	VNS	1.0	1.0	1.0	**1.0**	1.5	1.1	2.3	**1.6**	1.3	1.0	1.5	**1.3**
Sparta	Jumbo	1.1	1.0	1.0	**1.0**	1.0	1.1	2.3	**1.5**	1.0	1.0	2.2	**1.4**
Sparta	Ladino	1.1	1.0	1.0	**1.0**	1.0	2.1	2.7	**1.9**	1.3	1.0	2.2	**1.5**
Tekapo	Endura	1.1	1.0	1.0	**1.0**	1.3	1.0	1.0	**1.1**	1.3	2.0	3.3	**2.2**
Tekapo	VNS	1.3	1.2	1.2	**1.2**	1.3	1.2	2.3	**1.6**	1.1	2.0	2.5	**1.9**
Tekapo	Star Fire	1.5	1.0	1.0	**1.2**	1.5	2.5	3.7	**2.6**	1.1	2.0	2.8	**2.0**
Tekapo	Kopu II	1.6	1.0	1.0	**1.2**	1.1	1.1	2.6	**1.6**	1.6	3.0	4.0	**2.9**
Tekapo	Ladino	1.3	1.0	1.0	**1.1**	1.0	2.0	1.0	**1.3**	1.8	2.8	3.8	**2.8**
Aries	Endura	1.6	1.0	1.0	**1.2**	2.6	3.6	4.0	**3.4**	2.1	4.7	4.8	**3.9**
Aries	VNS	2.3	1.5	1.5	**1.8**	2.1	2.5	3.7	**2.8**	3.0	4.7	5.0	**4.2**
Aries	Star Fire	2.1	1.0	1.0	**1.4**	1.8	1.2	2.3	**1.8**	3.0	4.7	5.0	**4.2**
Aries	Kopu II	1.5	1.0	1.0	**1.2**	2.3	3.0	3.5	**2.9**	2.1	5.0	5.0	**4.0**
Aries	Ladino	1.5	1.0	1.0	**1.2**	1.6	2.3	2.7	**2.2**	1.8	5.0	5.0	**3.9**
Barfort	VNS	1.5	1.0	1.0	**1.2**	1.8	1.6	2.7	**2.0**	2.1	3.3	3.8	**3.1**
Barfort	Start	1.6	1.0	1.0	**1.2**	2.0	1.5	3.0	**2.2**	1.6	3.0	4.5	**3.0**
Barfort	Alice	1.6	1.0	1.0	**1.2**	2.0	2.3	2.7	**2.3**	1.3	4.3	5.0	**3.5**
Barfort	Ladino	1.3	1.0	1.0	**1.1**	1.8	2.3	2.8	**2.3**	1.0	4.3	5.0	**3.4**
Calibra	VNS	1.5	1.2	1.2	**1.3**	1.0	1.7	2.7	**1.8**	1.3	1.6	3.8	**2.2**
Calibra	Jumbo	1.1	1.0	1.0	**1.0**	1.6	1.8	1.0	**1.5**	1.1	3.3	4.7	**3.0**
Calibra	Ladino	1.6	1.0	1.0	**1.2**	1.6	3.1	2.3	**2.3**	1.0	3.0	4.2	**2.7**
Mara	VNS	1.3	1.0	1.0	**1.1**	1.8	2.0	1.0	**1.6**	1.5	3.0	3.0	**2.5**
Mara	Start	1.6	1.0	1.0	**1.2**	2.3	2.5	3.0	**2.6**	1.3	3.8	4.3	**3.1**
Mara	Alice	1.1	1.0	1.0	**1.0**	2.0	2.0	1.8	**1.9**	1.5	4.2	5.0	**3.6**

Table 2 continued

Grass Cultivars	Clover Cultivars	KBS '04	KBS '05	KBS '06	KBS AVE	LC '04	LC '05	LC '06	LC AVE	UPES '04	UPES '05	UPES '06	UPES AVE
Mara	Ladino	1.0	1.0	1.0	**1.0**	2.1	1.3	1.7	**1.7**	1.3	3.3	4.0	**2.9**
Mvck G	Endura	2.0	1.4	1.3	**1.6**	2.3	3.3	4.0	**3.2**	4.6	5.0	5.0	**4.9**
Mvck G	VNS	2.3	2.2	2.2	**2.2**	2.1	3.0	4.0	**3.0**	4.1	5.0	5.0	**4.7**
Mvck G	Star Fire	2.0	1.4	1.3	**1.6**	2.6	4.0	4.3	**3.6**	3.6	5.0	5.0	**4.5**
Mvck G	Kopu II	2.3	1.0	1.0	**1.4**	2.5	3.6	3.7	**3.3**	2.8	5.0	5.0	**4.3**
Mvck G	Ladino	2.1	1.0	1.0	**1.4**	2.8	3.0	4.3	**3.4**	2.8	5.0	5.0	**4.3**
Quartet	Endura	1.3	1.0	1.0	**1.1**	1.5	3.5	4.0	**3.0**	1.3	3.7	5.0	**3.3**
Quartet	VNS	2.0	1.0	1.0	**1.3**	1.6	3.1	4.3	**3.0**	2.0	3.7	5.0	**3.6**
Quartet	Ladino	1.5	1.0	1.0	**1.2**	1.5	1.1	3.0	**1.9**	2.0	5.0	5.0	**4.0**
Quartet	Star Fire	2.0	1.0	1.0	**1.3**	1.3	1.3	2.7	**1.8**	2.0	4.2	4.8	**3.7**
Quartet	Kopu II	2.0	1.0	1.0	**1.3**	1.5	1.8	3.0	**2.1**	2.0	4.5	4.7	**3.7**
Tonga	Endura	1.3	1.0	1.0	**1.1**	1.1	2.4	3.3	**2.3**	1.0	2.7	4.0	**2.6**
Tonga	VNS	1.6	1.0	1.0	**1.2**	1.0	2.4	3.7	**2.4**	1.3	3.0	4.7	**3.0**
Tonga	Star Fire	2.0	1.0	1.0	**1.3**	1.0	1.6	2.3	**1.6**	1.0	3.7	4.7	**3.1**
Tonga	Kopu II	1.3	1.0	1.0	**1.1**	1.1	1.0	1.7	**1.3**	1.1	4.0	4.7	**3.3**
Tonga	Ladino	1.5	1.0	1.0	**1.2**	1.1	1.1	2.7	**1.6**	1.3	5.0	5.0	**3.8**
Bronson	VNS	1.3	1.0	1.0	**1.1**	1.0	1.2	1.0	**1.1**	1.1	1.3	1.0	**1.1**
Bronson	Endura	1.5	1.0	1.0	**1.2**	1.0	1.2	1.0	**1.1**	1.0	2.3	1.0	**1.4**
Bronson	Kopu II	1.5	1.0	1.0	**1.2**	1.1	1.1	2.0	**1.4**	1.0	1.3	1.0	**1.1**
Bronson	Ladino	1.1	1.0	1.0	**1.0**	1.0	1.0	1.0	**1.0**	1.0	1.3	1.0	**1.1**
Bronson	Star Fire	1.1	1.0	1.0	**1.0**	1.1	1.3	1.3	**1.2**	1.3	1.6	1.0	**1.3**
K5666V	VNS	1.6	1.0	1.0	**1.2**	1.3	2.4	2.0	**1.9**	1.8	2.3	1.3	**1.8**
K5666V	Endura	1.1	1.0	1.0	**1.0**	1.5	2.4	1.7	**1.9**	1.6	2.0	1.0	**1.5**
K5666V	Kopu II	1.8	1.0	1.0	**1.3**	1.1	2.1	2.2	**1.8**	1.8	2.0	1.3	**1.7**
K5666V	Ladino	1.3	1.0	1.0	**1.1**	1.1	1.1	1.2	**1.1**	2.0	1.8	1.3	**1.7**
K5666V	Star Fire	1.3	1.0	1.0	**1.1**	1.6	1.0	1.0	**1.2**	1.5	2.0	1.7	**1.7**
Barolex	Alice	1.6	1.0	1.0	**1.2**	1.8	1.4	1.0	**1.4**	1.0	1.6	1.0	**1.2**
Barolex	VNS	1.5	1.0	1.0	**1.2**	1.0	1.0	1.3	**1.1**	1.3	1.6	1.3	**1.4**
Barolex	Ladino	1.3	1.0	1.0	**1.1**	1.0	1.0	1.3	**1.1**	1.1	1.8	1.0	**1.3**
Barolex	Start	1.5	1.0	1.0	**1.2**	1.3	1.7	2.0	**1.7**	1.3	1.5	1.3	**1.4**
	Mean	**1.5**	**1.0**	**1.0**		**1.5**	**1.8**	**2.4**		**1.6**	**2.9**	**3.3**	
	CV	**29**	**20**	**20**		**28**	**47**	**51**		**23**	**19**	**14**	
	LSD(0.05)	**0.7**	**0.3**	**0.4**		**0.7**	**0.0**	**0.0**		**0.6**	**0.9**	**0.8**	

1= less than 20% of the vegetation killed; 5= Greater than 80% of the vegetation was killed
Grass cultivars:
Festulolium (Duo and Hykor)
Orchardgrass (Amba, Niva, Sparta and Tekapo)
Perennial ryegrass (Aries, Barfort, Calibra, Mara, Maverick Gold, Quartet, and Tonga,)
Tall fescue (Bronson, K5666V, and Barolex)
Clover cultivars:
Red clover (Start Fire, Start and VNS); white clover (Ladino, Alice, Kopu II and Jumbo); Kura (Endura)

Table 3. The mean values of spring and fall ground cover rating of 16 grass cultivars established in monoculture at Kellogg Biological Research Station (KBS), Lake City (LC) and Upper Peninsula Experiment Station (UPES) over a three-year period.

	KBS						LC						UPES					
Grass	Spring	Fall	Spring	Fall	Spring	3-yr	Spring	Fall	Spring	Fall	Spring	3-yr	Spring	Fall	Spring	Fall	Spring	3-yr
Cultivars	2004	2004	2005	2005	2006	Ave	2004	2004	2005	2005	2006	Ave	2004	2004	2005	2005	2006	Ave
Duo	4.3	4.5	5.0	4.0	4.0	**4.4**	4.3	4.3	3.0	5.0	4.3	**4.2**	4.0	3.9	1.7	1.8	1.0	**2.5**
Hykor	4.3	4.6	5.0	5.0	5.0	**4.8**	5.0	5.0	4.8	5.0	5.0	**5.0**	5.0	4.4	5.0	5.0	3.8	**4.6**
Tekapo	4.0	4.6	4.7	4.8	4.7	**4.6**	4.1	4.6	2.3	4.7	4.7	**4.1**	4.6	4.3	2.7	5.0	1.0	**3.5**
Amba	4.1	4.3	4.8	3.7	4.7	**4.3**	4.5	4.6	4.0	5.0	4.0	**4.4**	5.0	3.9	3.7	5.0	3.0	**4.1**
Niva	4.3	4.6	5.0	5.0	4.7	**4.7**	4.6	4.5	3.5	5.0	4.3	**4.4**	5.0	4.3	3.7	5.0	2.7	**4.1**
Sparta	4.1	5.0	4.8	4.5	4.7	**4.6**	4.8	4.6	4.0	5.0	3.7	**4.4**	4.8	4.4	4.0	5.0	2.5	**4.1**
Aries	4.0	4.6	4.8	3.3	3.7	**4.1**	4.1	4.1	1.7	3.5	2.7	**3.2**	2.1	2.2	1.0	2.0	1.0	**1.7**
Mvrck Gold	3.5	3.3	3.0	1.6	1.3	**2.5**	1.8	3.6	2.0	3.5	3.7	**2.9**	2.3	1.7	1.0	1.2	1.0	**1.4**
Quartet	3.8	4.0	5.0	2.8	2.7	**3.7**	3.8	4.3	2.3	5.0	5.0	**4.1**	3.3	2.8	1.0	3.0	1.0	**2.2**
Tonga	4.1	4.6	5.0	2.7	3.3	**3.9**	3.5	4.1	2.3	5.0	4.0	**3.8**	4.1	3.9	1.7	2.4	1.0	**2.6**
Barfort	4.3	4.5	5.0	3.5	3.5	**4.2**	3.8	4.8	2.3	5.0	4.0	**4.0**	4.0	4.0	2.0	4.8	1.0	**3.2**
Mara	4.1	5.0	5.0	4.7	4.7	**4.7**	4.6	4.3	1.3	4.3	3.3	**3.6**	4.1	4.1	2.0	5.0	1.0	**3.2**
Calibra	3.6	4.6	5.0	3.7	4.0	**4.2**	3.6	4.1	2.3	5.0	5.0	**4.0**	4.3	4.1	2.0	5.0	1.0	**3.3**
Bronson	4.6	5.0	5.0	5.0	5.0	**4.9**	4.8	5.0	4.7	5.0	5.0	**4.9**	5.0	4.6	4.3	5.0	2.5	**4.3**
K5666V	4.1	5.0	5.0	4.8	4.3	**4.6**	4.6	5.0	3.7	5.0	5.0	**4.7**	4.8	4.3	2.7	5.0	1.0	**3.6**
Barolex	4.1	4.6	5.0	5.0	4.7	**4.7**	4.8	5.0	3.0	5.0	5.0	**4.6**	5.0	4.6	3.3	5.0	1.0	**3.8**
Mean	**4.1**	**4.5**	**4.9**	**4.0**	**4.1**		**4.3**	**4.6**	**3.0**	**4.8**	**4.3**		**4.2**	**3.8**	**2.6**	**4.1**	**1.6**	
CV	**7.1**	**9.6**	**4.8**	**10.5**	**21.2**		**14.6**	**7.2**	**23.2**	**10.5**	**13.4**		**6.4**	**10.9**	**15.3**	**6.1**	**14.3**	
LSD (0.05)	**0.4**	**0.7**	**0.4**	**0.7**	**1.4**		**1.0**	**0.5**	**1.1**	**0.8**	**0.9**		**0.5**	**0.7**	**0.7**	**0.5**	**0.4**	

1 = greater than 80 % of the vegetation missing; **5** = less than 20% of the vegetation missing

Grass cultivars:

Festulolium (Duo and Hykor)

Orchardgrass (Amba, Niva, Sparta and Tekapo)

Perennial ryegrass (Aries, Barfort, Calibra, Mara, Maverick Gold, Quartet, and Tonga,)

Tall fescue (Bronson, K5666V, and Barolex)

Table 4. The mean values of spring (S) and fall (F) ground cover rating of 67 binary mixtures treatments established at Kellogg Biological Research Station (KBS), Lake City (LC) and Upper Peninsula Experiment Station (UPES) over a three-year period (continued on the next pages).

		KBS						LC						UPES					
Grass	Clover	S	F	S	F	S	3-yr	S	F	S	F	S	3-yr	S	F	S	F	S	3-yr
Cultivars	Cultivars	'04	'04	'05	'05	'06	Ave	'04	'04	'05	'05	'06	Ave	'04	'04	'05	'05	'06	Ave
Duo	Endura	4.1	4.5	5.0	4.3	5.0	**4.6**	4.0	3.6	4.0	4.5	4.3	**4.1**	4.0	3.8	1.3	4.3	1.7	**3.0**
Duo	Star Fire	4.1	4.8	5.0	3.5	4.0	**4.3**	4.0	4.3	3.0	4.7	4.0	**4.0**	4.0	3.6	1.2	3.7	1.3	**2.8**
Duo	VNS	4.5	4.3	5.0	3.3	4.0	**4.2**	4.0	4.0	4.4	4.7	4.0	**4.2**	4.1	4.0	1.7	3.8	1.8	**3.1**
Duo	Ladino	4.3	4.6	5.0	3.0	3.5	**4.1**	4.6	4.0	3.7	4.5	4.0	**4.2**	4.3	4.4	1.2	3.7	2.0	**3.1**
Duo	Kopu II	4.1	5.0	4.9	2.3	2.8	**3.8**	4.8	4.0	4.4	4.7	3.7	**4.3**	4.6	4.2	1.0	4.0	2.3	**3.2**
Hykor	VNS	4.3	5.0	5.0	5.0	5.0	**4.9**	5.0	4.5	5.0	5.0	5.0	**4.9**	4.8	4.1	4.7	5.0	5.0	**4.7**
Hykor	Jumbo	4.3	4.3	5.0	5.0	5.0	**4.7**	5.0	4.5	4.4	5.0	5.0	**4.8**	5.0	4.1	4.3	5.0	5.0	**4.7**
Hykor	Ladino	4.3	4.3	5.0	5.0	5.0	**4.7**	5.0	4.6	5.0	5.0	5.0	**4.9**	5.0	4.2	4.3	5.0	4.7	**4.6**
Amba	VNS	4.3	4.6	5.0	4.2	3.8	**4.4**	4.6	4.3	5.0	5.0	4.3	**4.6**	5.0	4.3	5.0	5.0	5.0	**4.9**
Amba	Jumbo	4.3	4.0	4.5	3.7	4.7	**4.2**	4.3	4.0	4.5	5.0	4.7	**4.5**	5.0	4.3	4.5	5.0	4.3	**4.6**
Amba	Ladino	4.5	4.6	4.8	4.2	4.5	**4.5**	4.5	4.0	4.9	5.0	4.0	**4.5**	5.0	4.3	4.3	5.0	4.5	**4.6**
Niva	VNS	4.3	4.6	4.9	4.2	4.8	**4.6**	5.0	3.6	5.0	5.0	4.7	**4.7**	5.0	4.3	4.7	5.0	5.0	**4.8**
Niva	Jumbo	4.5	4.3	5.0	4.7	5.0	**4.7**	5.0	4.1	4.5	4.8	5.0	**4.7**	5.0	4.4	4.7	5.0	4.2	**4.7**
Niva	Ladino	4.1	4.6	5.0	4.3	4.8	**4.6**	4.8	4.6	3.9	5.0	4.3	**4.5**	5.0	4.2	5.0	5.0	4.3	**4.7**
Sparta	VNS	4.1	4.5	4.9	4.0	4.8	**4.5**	4.6	4.0	4.9	5.0	4.0	**4.5**	4.8	4.2	5.0	5.0	4.5	**4.7**
Sparta	Jumbo	4.6	5.0	5.0	4.3	4.8	**4.7**	5.0	4.3	4.9	4.8	4.7	**4.7**	5.0	4.2	5.0	4.8	3.7	**4.5**
Sparta	Ladino	4.3	5.0	5.0	4.3	4.8	**4.7**	5.0	4.3	3.5	4.8	4.3	**4.4**	5.0	4.2	4.7	5.0	3.8	**4.5**
Tekapo	Endura	4.1	4.6	5.0	4.8	5.0	**4.7**	4.8	3.6	5.0	5.0	4.7	**4.6**	4.5	4.0	3.7	5.0	2.8	**4.0**
Tekapo	VNS	4.5	4.8	4.9	4.0	4.2	**4.5**	4.6	4.1	4.5	4.8	4.7	**4.5**	4.1	4.4	3.7	5.0	3.5	**4.1**
Tekapo	Star Fire	4.0	4.6	4.9	4.8	4.0	**4.5**	4.8	4.0	2.5	4.5	4.7	**4.1**	4.0	4.1	3.5	4.8	3.0	**3.9**
Tekapo	Kopu II	4.1	4.6	4.7	4.2	4.4	**4.4**	4.5	3.8	4.0	5.0	4.3	**4.3**	4.1	4.6	2.3	5.0	2.0	**3.6**
Tekapo	Ladino	4.5	4.6	5.0	4.8	4.7	**4.7**	4.3	4.0	3.5	5.0	4.7	**4.3**	4.3	4.3	2.5	5.0	2.5	**3.7**

Table 4 continued

		KBS						LC						UPES					
Grass	Clover	S	F	S	F	S	3-yr	S	F	S	F	S	3-yr	S	F	S	F	S	3-yr
Cultivars	Cultivars	‘04	‘04	‘05	‘05	‘06	Ave	‘04	‘04	‘05	‘05	‘06	Ave	‘04	‘04	‘05	‘05	‘06	Ave
Aries	Endura	4.3	3.8	5.0	4.2	3.4	**4.1**	3.3	3.8	3.0	4.0	3.0	**3.4**	3.5	3.8	1.0	3.0	1.8	**2.6**
Aries	VNS	4.6	4.6	4.2	3.3	3.2	**4.0**	3.6	3.5	3.5	4.5	3.0	**3.6**	3.0	4.1	1.0	3.0	1.0	**2.4**
Aries	Star Fire	4.8	4.6	4.8	3.3	3.2	**4.1**	3.6	4.0	4.2	4.5	4.0	**4.1**	2.8	3.5	1.0	2.8	1.0	**2.2**
Aries	Kopu II	4.3	4.6	4.2	2.5	2.5	**3.6**	3.5	4.0	3.0	3.7	3.7	**3.6**	3.8	4.0	1.0	3.2	2.2	**2.8**
Aries	Ladino	4.5	4.3	4.8	3.0	3.4	**4.0**	4.0	4.0	3.4	4.4	3.3	**3.8**	3.8	3.9	1.0	3.1	2.7	**2.9**
Barfort	VNS	4.3	4.1	5.0	3.5	3.4	**4.1**	4.0	4.3	4.0	4.8	4.0	**4.2**	3.6	3.9	2.3	4.3	1.8	**3.2**
Barfort	Start	4.0	4.6	4.8	2.7	3.4	**3.9**	3.8	3.8	4.0	4.0	3.7	**3.9**	4.1	4.3	2.0	3.8	1.8	**3.2**
Barfort	Alice	4.1	4.6	4.8	2.7	2.7	**3.8**	3.8	4.0	3.7	4.8	4.0	**4.1**	4.6	4.3	1.0	4.3	3.2	**3.5**
Barfort	Ladino	4.3	5.0	5.0	3.0	2.3	**3.9**	3.6	4.0	3.0	5.0	4.3	**4.0**	4.5	4.2	1.0	4.2	2.5	**3.3**
Calibra	VNS	4.1	4.3	5.0	3.5	4.2	**4.2**	4.0	4.5	3.3	5.0	3.8	**4.1**	4.3	4.2	3.0	5.0	2.0	**3.7**
Calibra	Jumbo	4.3	4.6	5.0	3.5	3.8	**4.2**	4.1	4.0	3.0	4.8	4.3	**4.0**	4.8	4.1	2.0	4.0	2.0	**3.4**
Calibra	Ladino	4.6	5.0	5.0	3.2	3.4	**4.2**	4.0	4.1	3.0	4.8	4.7	**4.1**	5.0	4.3	2.3	4.5	2.7	**3.8**
Mara	VNS	4.3	4.6	5.0	4.7	4.7	**4.7**	4.0	4.0	3.7	4.8	4.0	**4.1**	4.0	4.1	2.3	4.8	2.8	**3.6**
Mara	Start	4.0	4.8	5.0	4.5	4.8	**4.6**	4.1	4.1	3.5	4.8	3.7	**4.0**	4.3	4.2	2.0	5.0	2.0	**3.5**
Mara	Alice	4.3	4.6	4.9	4.5	4.8	**4.6**	3.6	4.1	3.4	5.0	4.0	**4.0**	4.5	4.2	1.0	4.3	2.2	**3.2**
Mara	Ladino	4.3	4.8	5.0	4.3	4.5	**4.6**	3.6	4.3	4.0	4.8	3.7	**4.1**	4.5	4.2	1.7	4.0	2.0	**3.3**
MvckGd	Endura	3.8	3.3	4.2	2.7	2.5	**3.3**	3.8	3.1	3.3	3.8	3.3	**3.5**	1.3	3.0	1.0	2.2	1.5	**1.8**
MvckGd	VNS	4.0	4.0	4.2	2.1	2.3	**3.3**	4.3	4.1	3.0	3.2	3.3	**3.6**	2.3	2.9	1.0	2.2	1.0	**1.9**
MvckGd	Star Fire	4.1	3.5	4.4	2.7	2.0	**3.3**	3.1	2.8	2.3	3.4	4.3	**3.2**	2.3	3.0	1.0	3.0	1.5	**2.2**
MvckGd	Kopu II	3.5	4.0	3.9	2.5	2.3	**3.2**	3.3	3.3	3.0	3.5	3.7	**3.4**	3.0	3.7	1.0	4.0	3.2	**3.0**
MvckGd	Ladino	3.8	4.1	4.9	2.3	1.7	**3.4**	2.8	2.6	3.0	3.4	4.0	**3.2**	3.1	3.6	1.0	4.0	3.0	**2.9**
Quartet	Endura	4.1	4.1	5.0	3.5	4.4	**4.2**	4.0	4.5	2.5	5.0	4.3	**4.1**	4.0	3.6	1.7	3.8	1.0	**2.8**
Quartet	VNS	4.1	4.0	4.5	3.2	**2.2**	**3.6**	4.3	3.6	2.5	4.8	4.7	**4.0**	3.6	4.2	1.3	4.0	1.0	**2.8**
Quartet	Ladino	4.1	4.6	4.9	3.0	3.5	**4.0**	4.5	3.6	3.5	4.3	4.0	**4.0**	4.0	4.1	1.0	3.8	2.7	**3.1**
Quartet	Star Fire	4.1	4.3	4.5	3.3	3.4	**3.9**	4.3	3.6	3.4	4.5	4.0	**4.0**	3.6	4.3	2.1	4.0	1.7	**3.1**
Quartet	Kopu II	4.1	4.6	4.5	2.3	**1.7**	**3.4**	4.3	3.6	2.9	3.8	4.3	**3.8**	4.0	4.2	1.0	3.7	2.7	**3.1**

Table 4 continued

		KBS						LC						UPES					
Grass	**Clover**	S	F	S	F	S	**3-yr**	S	F	S	F	S	**3-yr**	S	F	S	F	S	**3-yr**
Cultivars	**Cultivars**	**'04**	**'04**	**'05**	**'05**	**'06**	**Ave**	**'04**	**'04**	**'05**	**'05**	**'06**	**Ave**	**'04**	**'04**	**'05**	**'05**	**'06**	**Ave**
Tonga	Endura	4.0	3.8	5.0	3.8	4.7	**4.3**	4.1	4.1	3.4	4.4	3.7	**3.9**	4.8	3.7	2.7	4.0	2.8	**3.6**
Tonga	VNS	4.0	4.0	4.9	3.2	4.2	**4.1**	4.5	4.5	2.7	5.0	4.8	**4.3**	4.1	4.3	2.3	4.5	1.7	**3.4**
Tonga	Star Fire	4.0	3.6	4.4	2.7	2.7	**3.5**	4.5	4.1	3.4	5.0	4.7	**4.3**	4.5	4.1	2.0	4.3	1.8	**3.3**
Tonga	Kopu II	4.5	4.6	4.5	2.7	2.7	**3.8**	4.1	4.0	4.4	4.8	4.0	**4.3**	4.3	4.0	1.0	4.0	2.0	**3.1**
Tonga	Ladino	4.3	4.6	5.0	2.8	3.2	**4.0**	4.5	4.3	4.5	3.5	4.3	**4.2**	4.6	4.5	1.0	3.7	2.7	**3.3**
Bronson	VNS	4.6	4.6	5.0	5.0	5.0	**4.8**	5.0	5.0	4.7	5.0	5.0	**4.9**	4.5	4.4	4.3	5.0	5.0	**4.6**
Bronson	Endura	4.5	4.6	5.0	5.0	5.0	**4.8**	5.0	4.6	4.7	5.0	5.0	**4.9**	4.3	4.0	3.0	4.8	5.0	**4.2**
Bronson	Kopu II	4.8	4.8	4.8	5.0	4.8	**4.8**	4.8	5.0	5.0	4.8	5.0	**4.9**	4.6	4.4	3.8	5.0	5.0	**4.6**
Bronson	Ladino	4.8	5.0	5.0	5.0	5.0	**5.0**	4.6	4.1	5.0	5.0	4.7	**4.7**	4.6	4.4	4.3	5.0	5.0	**4.7**
Bronson	Star Fire	4.1	4.6	5.0	5.0	4.7	**4.7**	4.5	4.6	5.0	5.0	5.0	**4.8**	4.3	4.4	4.0	5.0	5.0	**4.5**
K5666V	VNS	4.1	5.0	4.9	4.7	4.8	**4.7**	4.6	4.6	3.4	4.7	5.0	**4.5**	4.0	4.1	3.0	5.0	4.7	**4.2**
K5666V	Endura	4.3	4.6	5.0	5.0	5.0	**4.8**	4.3	4.1	3.5	5.0	5.0	**4.4**	4.0	4.1	3.3	4.8	5.0	**4.2**
K5666V	Kopu II	4.1	5.0	4.9	4.7	4.2	**4.6**	4.6	4.8	3.5	5.0	4.7	**4.5**	4.1	4.0	3.5	5.0	4.7	**4.3**
K5666V	Ladino	4.6	5.0	5.0	4.3	4.8	**4.7**	4.5	4.0	4.7	5.0	5.0	**4.6**	4.1	4.1	3.7	5.0	4.7	**4.3**
K5666V	Star Fire	4.3	4.3	5.0	4.8	4.5	**4.6**	4.1	4.5	5.0	5.0	5.0	**4.7**	4.1	4.3	3.3	5.0	4.2	**4.2**
Barolex	Alice	4.0	4.8	5.0	4.2	4.4	**4.5**	4.6	4.6	4.7	5.0	5.0	**4.8**	4.8	4.1	3.7	5.0	5.0	**4.5**
Barolex	VNS	4.0	5.0	5.0	5.0	4.8	**4.8**	5.0	4.6	4.7	5.0	5.0	**4.9**	4.5	4.1	3.7	5.0	4.7	**4.4**
Barolex	Ladino	4.0	5.0	5.0	5.0	4.7	**4.7**	4.6	4.5	4.7	5.0	5.0	**4.8**	4.6	4.0	3.7	5.0	5.0	**4.5**
Barolex	Start	4.1	5.0	4.9	5.0	4.7	**4.7**	4.5	4.6	4.4	4.5	5.0	**4.6**	4.1	4.5	3.7	5.0	4.3	**4.3**
	Mean	**4.2**	**4.5**	**4.8**	**3.8**	**3.9**		**4.3**	**4.1**	**3.4**	**4.1**	**2.5**		**4.2**	**4.1**	**2.6**	**4.4**	**3.1**	
	CV	**9.1**	**10.3**	**5.9**	**15.4**	**15.5**		**10.5**	**12.9**	**0.0**	**0.0**	**0.0**		**8.4**	**8.3**	**20.6**	**9.4**	**21.2**	
	LSD (0.05)	**0.6**	**0.7**	**0.4**	**0.9**	**1.0**		**0.7**	**0.8**	**0.0**	**0.0**	**0.0**		**0.6**	**0.6**	**0.9**	**0.7**	**1.1**	

1 = greater than 80 % of the vegetation missing; **5** = less than 20% of the vegetation missing

Grass cultivars: Festulolium (Duo and Hykor); Orchardgrass (Amba, Niva, Sparta and Tekapo); Perennial ryegrass (Aries, Barfort, Calibra, Mara, Maverick Gold, Quartet, and Tonga,), Tall fescue (Bronson, K5666V, and Barolex)

Clover cultivars: Red clover (Start Fire, Start and VNS), white clover (Ladino, Alice, kopu II, and Jumbo), and Kura (Endura)

Table 5. The mean values of winter injury rating of eight clover cultivars established in monoculture at Kellogg Biological Research Station (KBS), Lake City (LC) and Upper Peninsula Experiment Station (UPES) over a three-year period

Clover Species	Clover Cultivars	KBS				*LC			#UPES		
		2004	2005	2006	Ave	2004	2005	2006	2004	2005	Ave
Kura clover	Endura	1.0	1.0	1.0	**1.0**	1.5	1.0	1.0	3.8	1.0	**2.4**
	Star Fire	3.1	2.0	2.0	**2.4**	1.8	NA	NA	2.6	1.0	**1.8**
Red Clover	Start	2.8	1.5	1.5	**1.9**	1.5	NA	NA	2.6	1.0	**1.8**
	VNS	3.0	1.8	2.0	**2.3**	1.1	NA	NA	2.5	1.0	**1.8**
	Kopu II	2.3	1.3	1.4	**1.7**	1.0	NA	NA	2.0	1.0	**2.0**
White Clover	Alice	2.1	1.0	1.0	**1.4**	1.0	NA	NA	1.0	1.0	**1.0**
	Jumbo	2.0	1.5	1.5	**1.7**	1.0	NA	NA	1.6	1.0	**1.3**
	Ladino	2.5	1.0	1.0	**1.5**	1.0	NA	NA	1.6	1.0	**1.3**
	Mean	**2.4**	**1.4**	**1.4**		**1.25**	**1.0**	**1.0**	**2.2**	**1.0**	
	CV	**18.9**	**23.9**	**23.4**		**0.0**	**0.0**	**0.0**	**26.8**	**0.0**	
	LSD (0.05)	**0.8**	**0.6**	**0.6**		**0.6**	**NS**	**NS**	**1.05**	**NS**	

1= less than 20% of the vegetation killed.
5= Greater than 80% of the vegetation was killed.
* LC 2005 and 2006 white and red clovers data are not included.
# UPES 2006 data are not included.
NA= Not applicable.

Table 6. The mean values of spring and fall ground cover rating of eight clover cultivars established in monoculture at Kellogg Biological Research Station (KBS), Lake City (LC) and Upper Peninsula Experiment Station (UPES) over a two-year period.

		KBS				*LC					UPES				
Clover	**Clover**	**spring**	**fall**	**spring**		**spring**	**fall**	**spring**	**fall**		**spring**	**fall**	**spring**	**fall**	
Species	**Cultivars**	2004	2004	2005	**Ave**	2004	2004	2005	2005	**Ave**	2004	2004	2005	2005	**Ave**
Kura clover	Endura	4.1	4.0	4.0	**4.0**	4.2	1.0	3.2	2.7	**2.8**	1.5	4.4	2.0	2.3	**2.6**
Red	Star Fire	3.5	3.1	3.5	**3.4**	4.2	2.0	NA	NA	**3.1**	3.0	1.3	1.3	1.3	**1.7**
Clover	Start	4.0	3.8	3.5	**3.8**	4.2	2.7	NA	NA	**3.5**	3.1	2.0	1.0	1.7	**2.0**
	VNS	3.6	3.6	3.5	**3.6**	4.3	2.7	NA	NA	**3.5**	2.5	1.4	1.0	1.8	**1.7**
	Kopu II	3.8	4.6	4.2	**4.2**	4.8	3.2	NA	NA	**4.0**	4.0	4.0	3.7	4.3	**4.0**
White	Alice	4.3	4.3	4.7	**4.4**	5.0	3.0	NA	NA	**4.0**	4.6	2.7	4.7	4.7	**4.2**
Clover	Jumbo	4.0	4.6	4.0	**4.2**	5.0	2.5	NA	NA	**3.8**	4.0	3.1	4.0	3.8	**3.7**
	Ladino	3.6	5.0	3.8	**4.1**	5.0	3.5	NA	NA	**4.3**	4.0	4.1	3.7	4.5	**4.1**
	Mean	**3.8**	**4.1**	**3.89**		**4.6**	**2.6**	**0.0**	**0.0**		**3.3**	**2.9**	**2.7**	**3.1**	
	CV	**10.4**	**12.4**	**12.3**		**8.1**	**16.5**	**0.0**	**0.0**		**14.2**	**36.1**	**15.3**	**22.8**	
	LSD (0.05)	**0.7**	**0.9**	**0.8**		**0.6**	**0.7**	**0.0**	**0.0**		**0.8**	**1.8**	**0.7**	**1.2**	

1 = greater than 80 % of the vegetation missing
5 = less than 20% of the vegetation missing
* LC 2005 data included kura clover only.
KBS fall of 2005 data are not included.

Table 7. The mean values of spring (S) and fall (F) percentage clover rating of festulolium-clover binary mixtures established at Kellogg Biological Research Station (KBS), Lake City (LC) and Upper Peninsula Experiment Station (UPES) over a three -year period.

		KBS						*LC					UPES					
Fest.	Clover	S'04	F'04	S'05	F'05	S'06	3-yr ave	S'04	F'04	S'05	F'05	2-yr ave	S'04	F'04	S'05	F'05	S'06	3-yr ave
Duo	Endura	4.3	4.0	5.0	4.5	5.0	**4.6**	2.1	1.6	2.0	3.8	**2.4**	1.6	2.1	2.0	2.2	1.7	**1.9**
Duo	Star Fire	4.6	4.1	4.2	1.3	1.3	**3.1**	2.6	1.6	1.7	4.4	**2.6**	3.1	3.6	2.0	3.3	1.3	**2.7**
Duo	VNS	4.8	4.0	4.2	1.2	1.7	**3.2**	3.3	2.1	2.0	4.0	**2.9**	3.1	2.4	1.0	2.7	1.0	**2.0**
Duo	Ladino	4.6	4.6	5.0	1.2	1.3	**3.3**	4.0	1.8	3.0	4.5	**3.3**	4.0	3.2	4.0	3.8	2.8	**3.6**
Duo	Kopu II	4.5	5.0	5.0	3.0	1.3	**3.8**	3.3	1.6	1.0	2.5	**2.1**	4.0	3.5	3.3	3.8	2.3	**3.4**
Hykor	VNS	4.1	3.0	3.5	1.0	1.0	**2.5**	2.0	1.3	1.4	2.0	**1.7**	2.3	2.2	1.0	2.0	1.0	**1.7**
Hykor	Jumbo	4.6	3.8	4.0	1.0	1.0	**2.9**	2.3	1.0	1.0	1.7	**1.5**	3.8	2.8	2.7	1.0	1.0	**2.3**
Hykor	Ladino	4.5	4.3	4.7	1.0	1.0	**3.1**	1.6	1.0	1.0	1.5	**1.3**	3.3	2.4	2.7	2.7	1.0	**2.4**
	Mean	**4.5**	**4.2**	**4.4**	**1.7**	**1.5**		**2.4**	**1.4**	**1.4**	**2.8**		**3.3**	**2.6**	**2.1**	**2.5**	**1.5**	
	CV	**11**	**13**	**10**	**39**	**25**		**30**	**34**	**45**	**25**		**14**	**33**	**29**	**30**	**31**	
	LSD(0.05)	**0.7**	**0.8**	**0.7**	**1.0**	**0.6**		**1.1**	**0.8**	**NS**	**NS**		**0.8**	**1.3**	**1.0**	**1.2**	**0.8**	

Mean, LSD and CV values represent the analysis of the entire 67 binary mixtures treatments.

1= Less than 20% of the clover stand appears in the mixture; **5** = 80% or greater of the mixture is cover with the clover stand
* LC 2006 data are not included
Clover Cultivar: Red clover (Star Fire, VNS and Start); White clover (Kopu II, Ladino and Jumbo); Kura (Endura)

Table 8. The mean values of spring (S) and fall (F) percentage clover rating of orchardgrass-clover binary mixtures established at Kellogg Biological Research Station (KBS), Lake City (LC) and Upper Peninsula Experiment Station (UPES) over a three-year period.

		KBS						*LC					UPES					
Orchardgrass Cultivars	**Clover Cultivars**	S04	F04	S05	F05	S06	**3-yr ave**	S04	F04	S05	F05	**2-yr ave**	S04	F04	S05	F05	S06	**3-yr ave**
Amba	VNS	4.0	4.0	4.3	1.5	1.0	**3.0**	1.8	1.6	1.0	2.4	**1.7**	2.5	3.0	1.0	1.2	1.0	**1.7**
Amba	Jumbo	4.5	4.3	5.0	1.3	1.0	**3.2**	2.0	1.6	1.4	2.0	**1.8**	3.1	2.0	2.0	1.3	1.0	**1.8**
Amba	Ladino	4.5	5.0	5.0	1.0	1.0	**3.3**	2.0	1.1	1.0	1.9	**1.5**	3.6	1.8	2.0	1.5	1.0	**2.0**
Niva	VNS	4.0	3.8	4.5	1.0	1.0	**2.9**	1.8	1.3	1.0	3.2	**1.8**	2.6	2.5	1.0	1.7	1.0	**1.8**
Niva	Jumbo	4.3	4.0	5.0	1.0	1.0	**3.1**	1.0	1.0	1.0	1.7	**1.2**	4.1	2.4	3.0	1.7	1.0	**2.5**
Niva	Ladino	4.5	4.5	5.0	1.0	1.0	**3.2**	2.0	1.3	1.1	3.4	**2.0**	4.0	1.9	2.0	1.3	1.0	**2.1**
Sparta	VNS	4.0	4.0	4.8	1.0	1.0	**3.0**	1.6	1.1	1.1	2.1	**1.5**	2.8	1.2	1.0	1.7	1.0	**1.5**
Sparta	Jumbo	4.5	5.0	5.0	1.8	1.7	**3.6**	1.6	1.1	1.6	2.6	**1.7**	4.5	3.1	3.0	1.7	1.0	**2.6**
Sparta	Ladino	4.6	4.6	4.5	1.3	1.7	**3.3**	2.1	1.5	1.4	3.2	**2.1**	4.3	2.3	2.0	1.7	1.0	**2.3**
Tekapo	Endura	4.1	4.3	4.7	4.0	4.3	**4.3**	2.6	1.0	1.4	2.7	**1.9**	2.0	1.0	2.0	1.7	2.0	**1.7**
Tekapo	VNS	4.1	3.1	4.2	1.0	1.3	**2.7**	2.6	1.0	1.9	2.9	**2.1**	2.0	1.5	1.0	1.8	1.0	**1.5**
Tekapo	Star Fire	3.8	3.5	4.2	1.0	1.0	**2.7**	2.0	1.3	1.0	3.2	**1.9**	2.3	2.3	1.0	1.7	1.0	**1.7**
Tekapo	Kopu II	4.6	4.3	5.0	1.7	1.0	**3.3**	1.8	1.1	1.0	1.9	**1.5**	3.8	1.8	2.0	1.5	1.0	**2.0**
Tekapo	Ladino	4.5	5.0	5.0	1.0	1.0	**3.3**	2.5	1.3	1.0	2.5	**1.8**	4.0	1.4	2.0	1.2	1.0	**1.9**
	Mean	**4.4**	**4.2**	**4.4**	**1.7**	**1.5**		**2.4**	**1.4**	**1.4**	**2.8**		**3.3**	**2.6**	**2.1**	**2.5**	**1.5**	
	CV	**11**	**13**	**10**	**39**	**25**		**30**	**34**	**45**	**25**		**14**	**33**	**29**	**30**	**31**	
	LSD(0.05)	**0.7**	**0.8**	**0.7**	**1.0**	**0.6**		**1.1**	**0.8**	**NS**	**NS**		**0.8**	**1.38**	**1.0**	**1.2**	**0.8**	

Mean LSD and CV values represent the analysis of the entire 67 binary mixtures treatments.

1= Less than 20% of the clover stand appears in the mixture
5 = 80% or greater of the mixture is cover with the clover stand
* LC 2006 data are not included

Clover Cultivars: Red clover (Star Fire, VNS and Start); White clover (Kopu II, Ladino and Jumbo); Kura (Endura)

Table 9. The mean values of spring (S) and fall (F) percentage clover of perennial ryegrass-clover mixtures in three locations (continued on the next page)

		KBS						LC					UPES					
P. ryegrss	Clover	S04	F04	S05	F05	S06	3-yr ave	S04	F04	S05	F05	2-yr ave	S04	F04	S05	F05	S06	3-yr ave
Aries	Endura	3.8	3.5	4.8	3.5	4.0	**3.9**	2.1	1.0	2.0	2.8	**2.0**	2.0	2.0	2.0	3.0	2.2	**2.2**
Aries	VNS	4.0	3.8	4.3	1.3	1.0	**2.9**	3.1	1.8	1.9	3.4	**2.6**	3.0	2.7	1.0	2.0	1.0	**1.9**
Aries	Star Fire	4.5	4.1	4.2	1.7	1.0	**3.1**	2.6	1.6	1.9	3.6	**2.4**	3.0	3.4	2.0	3.3	1.3	**2.7**
Aries	Kopu II	4.6	5.0	4.5	2.2	1.0	**3.5**	2.0	1.6	2.0	3.8	**2.4**	3.6	2.2	2.0	3.3	2.2	**2.7**
Aries	Ladino	4.8	5.0	5.0	1.7	1.3	**3.6**	2.6	1.6	2.0	2.8	**2.3**	4.0	1.8	2.0	3.8	3.0	**3.0**
Barfort	VNS	4.6	4.0	4.3	1.0	1.3	**3.0**	3.1	2.0	2.4	4.0	**2.9**	3.3	2.7	1.0	2.2	1.0	**2.0**
Barfort	Start	4.8	4.8	4.8	1.7	1.3	**3.5**	2.5	1.8	1.9	3.4	**2.4**	3.5	4.0	2.0	2.3	1.0	**2.5**
Barfort	Alice	5.0	5.0	5.0	2.0	1.7	**3.7**	3.1	1.5	1.4	3.4	**2.4**	4.5	3.9	4.0	4.5	3.3	**4.1**
Barfort	Ladino	4.8	5.0	5.0	1.3	1.0	**3.4**	2.5	1.6	1.0	4.0	**2.3**	4.1	4.0	4.0	4.0	3.0	**3.8**
Calibra	VNS	4.1	4.0	4.0	1.3	1.3	**2.9**	3.5	2.0	1.6	3.4	**2.6**	3.0	3.1	1.0	2.5	1.0	**2.1**
Calibra	Jumbo	4.8	5.0	5.0	1.8	1.7	**3.7**	2.5	1.1	1.0	3.7	**2.1**	4.1	2.4	4.0	3.0	2.0	**3.1**
Calibra	Ladino	4.8	5.0	5.0	2.3	1.3	**3.7**	2.3	1.1	1.4	3.2	**2.0**	4.0	3.4	3.0	3.0	1.7	**3.1**
Mara	VNS	4.1	3.3	3.5	1.2	1.0	**2.6**	2.3	2.1	1.7	3.7	**2.5**	2.6	1.6	1.0	1.7	1.0	**1.6**
Mara	Start	4.0	4.3	3.3	1.0	1.0	**2.7**	2.0	2.5	1.4	2.2	**2.0**	3.1	3.7	1.0	2.2	1.0	**2.3**
Mara	Alice	4.8	4.6	3.8	1.3	1.0	**3.1**	2.3	1.5	1.0	2.7	**1.9**	4.3	2.6	4.0	3.3	1.8	**3.3**
Mara	Ladino	4.5	4.8	4.2	1.0	1.0	**3.1**	1.6	1.0	1.0	2.4	**1.5**	4.0	3.3	3.0	3.2	1.5	**2.9**
Mvrck Gld	Endura	4.5	3.6	4.5	3.8	5.0	**4.3**	3.0	1.0	2.4	3.2	**2.4**	3.6	1.7	2.0	3.5	2.5	**2.7**
Mvrck Gld	VNS	4.3	4.0	3.7	1.3	1.3	**2.9**	4.0	1.1	1.7	3.4	**2.6**	4.3	4.3	1.0	3.5	1.3	**2.9**
Mvrck Gld	Star Fire	4.8	4.0	3.8	1.5	1.3	**3.1**	3.3	2.0	1.0	3.7	**2.5**	4.3	3.7	2.0	4.3	1.7	**3.2**
Mvrck Gld	Kopu II	4.3	4.6	4.7	2.3	1.3	**3.4**	2.6	1.1	1.7	3.0	**2.1**	4.6	4.0	4.0	5.0	4.2	**4.4**
Mvrck Gld	Ladino	4.5	4.6	4.7	1.7	1.0	**3.3**	3.1	1.1	1.0	3.7	**2.2**	4.8	4.4	5.0	5.0	4.0	**4.6**
Quartet	Endura	4.3	4.3	5.0	3.8	5.0	**4.5**	2.8	1.1	1.4	3.2	**2.1**	1.8	1.6	2.0	2.2	1.7	**1.9**
Quartet	VNS	4.3	4.1	4.2	1.3	1.3	**3.0**	3.5	2.5	1.4	4.4	**3.0**	3.1	3.5	1.0	2.0	1.0	**2.1**
Quartet	Ladino	5.0	5.0	5.0	2.8	1.0	**3.8**	4.1	1.5	2.4	2.7	**2.7**	4.1	2.3	3.0	4.2	3.0	**3.4**

Table 9 continued

P. ryegrss	Clover	KBS						LC					UPES					
		S04	F04	S05	F05	S06	3-yr ave	S04	F04	S05	F05	2-yr ave	S04	F04	S05	F05	S06	3-yr ave
Quartet	Star Fire	4.6	3.6	4.5	1.7	1.7	**3.2**	3.1	1.6	2.4	4.4	**2.9**	3.5	3.1	2.0	4.2	2.0	**3.0**
Quartet	Kopu II	4.8	4.6	5.0	3.0	1.7	**3.8**	3.1	1.3	2.4	2.9	**2.4**	4.1	2.6	2.0	3.0	2.0	**2.8**
Tonga	Endura	4.6	4.0	5.0	4.0	5.0	**4.5**	2.5	1.5	2.0	4.0	**2.5**	1.8	1.6	2.0	3.3	3.0	**2.4**
Tonga	VNS	4.6	3.6	4.3	1.3	1.0	**3.0**	3.5	1.0	1.1	4.3	**2.5**	3.3	3.2	1.0	2.5	1.0	**2.2**
Tonga	Star Fire	4.8	4.0	5.0	2.2	1.0	**3.4**	2.8	2.3	1.7	4.1	**2.7**	3.0	4.0	2.0	3.7	1.5	**2.8**
Tonga	Kopu II	5.0	5.0	5.0	2.7	1.7	**3.9**	2.8	1.0	1.0	3.0	**2.0**	4.0	2.4	3.0	3.2	2.7	**3.1**
Tonga	Ladino	5.0	5.0	5.0	2.8	1.7	**3.9**	3.1	1.6	1.4	3.5	**2.4**	4.3	3.1	4.0	3.7	3.2	**3.7**
	Mean	**4.4**	**4.2**	**4.4**	**1.7**	**1.5**		**2.4**	**1.4**	**1.4**	**2.8**		**3.3**	**2.6**	**2.1**	**2.5**	**1.5**	
	CV	**11**	**13**	**10**	**39**	**25**		**30**	**34**	**45**	**25**		**14**	**33**	**29**	**30**	**31**	
	LSD(0.05)	**0.7**	**0.8**	**0.7***	**1.0**	**0.6**		**1.1**	**0.8**	**NS**	**NS**		**0.8**	**1.4**	**1.0**	**1.2**	**0.8**	

Mean LSD and CV values represent the analysis of the entire 67 binary mixtures treatments.

1= Less than 20% of the clover stand appears in the mixture
5 = 80% or greater of the mixture is cover with the clover stand
* LC 2006 data are not included

Clover Cultivars: Red clover (Star Fire, VNS and Start); white clover (Kopu II, Ladino, Alice and Jumbo); Kura (Endura)

Table 10. The mean values of spring (S) and fall (F) percentage clover rating of tall fescue-clover binary mixtures established at Kellogg Biological Research (KBS), Lake City (LC) and Upper Peninsula Experiment Station (UPES) over a three-year period.

T. fescue	Clover	KBS						LC*					UPES					
Cultivars	Cultivars	S04	F04	S05	F05	S06	3-yr ave	S04	F04	S05	F05	2-yr ave	S04	F04	S05	F05	S06	3-yr ave
Bronson	VNS	3.8	3.6	3.3	1.2	1.0	**2.6**	1.5	1.0	1.4	2.2	**1.5**	2.5	2.6	1.0	1.3	1.0	**1.7**
Bronson	Endura	3.3	3.3	3.5	2.0	1.7	**2.8**	1.6	1.0	1.0	1.7	**1.3**	1.5	1.8	1.0	1.8	1.7	**1.6**
Bronson	Kopu II	3.8	4.6	4.0	1.2	1.0	**2.9**	2.0	1.0	1.0	1.4	**1.4**	3.5	2.6	2.0	1.3	1.0	**2.1**
Bronson	Ladino	4.8	4.6	4.8	1.0	1.0	**3.2**	2.0	1.0	1.0	1.7	**1.4**	3.5	2.2	2.0	2.3	1.0	**2.2**
Bronson	Star Fire	3.6	3.1	3.0	1.3	1.0	**2.4**	2.1	1.3	1.7	2.2	**1.8**	3.0	3.5	2.0	2.2	1.0	**2.3**
K5666V	VNS	4.1	3.0	3.0	2.3	1.0	**2.7**	2.3	1.0	1.0	2.0	**1.6**	2.5	1.6	1.0	1.8	1.0	**1.6**
K5666V	Endura	4.5	3.0	4.7	3.2	4.0	**3.9**	1.8	1.0	1.4	2.7	**1.7**	1.8	1.0	2.0	2.0	1.3	**1.6**
K5666V	Kopu II	4.6	5.0	5.0	1.3	1.3	**3.4**	2.0	1.3	1.0	2.7	**1.8**	3.3	1.2	2.0	1.5	1.0	**1.8**
K5666V	Ladino	4.8	5.0	5.0	1.2	1.0	**3.4**	2.2	1.2	1.0	1.9	**1.6**	3.3	2.3	2.0	1.3	1.0	**2.0**
K5666V	Star Fire	5.0	4.3	4.0	1.2	1.0	**3.1**	2.5	1.3	1.7	2.7	**2.1**	3.0	2.9	1.0	2.0	1.0	**2.0**
Barolex	Alice	3.8	3.6	4.5	1.0	1.0	**2.8**	1.5	1.1	1.0	1.5	**1.3**	3.5	2.9	3.0	1.8	1.0	**2.4**
Barolex	VNS	4.8	4.0	2.7	1.0	1.0	**2.7**	2.1	1.3	1.4	1.7	**1.6**	2.3	2.2	1.0	1.7	1.0	**1.6**
Barolex	Ladino	4.8	4.0	4.5	1.0	1.0	**3.1**	1.6	1.0	1.0	1.7	**1.3**	3.1	1.4	2.0	1.5	1.0	**1.7**
Barolex	Start	3.8	3.1	2.7	1.0	1.0	**2.3**	2.0	1.6	1.0	2.0	**1.7**	3.1	3.3	2.0	1.7	1.0	**2.2**
	Mean	**4.4**	**4.2**	**4.4**	**1.7**	**1.5**		**2.4**	**1.4**	**1.4**	**2.8**		**3.3**	**2.6**	**2.1**	**2.5**	**1.5**	
	CV	**11**	**13**	**10**	**39**	**25**		**30**	**34**	**45**	**25**		**14**	**33**	**29**	**30**	**31**	
	LSD (0.05)	**0.7**	**0.9**	**0.7**	**1.1**	**0.6**		**1.2**	**0.8**	**NS**	**NS**		**0.8**	**1.4**	**1.0**	**1.2**	**0.8**	

Mean, LSD and CV values represent the analysis of the entire 67 binary mixtures treatments.
1= Less than 20% of the clover stand appears in the mixture
5 = 80% or greater of the mixture is cover with the clover stand
* LC 2006 data are not included

Clover Cultivar: Red clover (Star Fire, VNS and Start); White clover (Kopu II, Ladino and Jumbo); Kura (Endura)

Table 11. Dry matter yield (tons acre^{-1}) of 16 grass cultivars established in monoculture at Kellogg Biological Research (KBS) over a two-year period.

Grass Species	/ Cultivars	2004 24-May	2004 24-Jun	2004 26-Jul	2004 9-Sep	2005 4-May	2005 14-Jun	2005 2-Aug	2005 13-Oct	Total 2-yr
		DM tons acre^{-1}								
Fest	Duo	0.75	0.52	0.27	0.29	0.41	0.16	0.31	0.13	**2.84**
	Hykor	1.08	0.84	0.82	0.39	1.16	0.56	0.51	0.30	**5.66**
OR	Tekapo	1.04	0.56	0.61	0.27	0.60	0.73	0.50	0.26	**4.57**
	Amba	0.85	0.82	0.46	0.20	0.88	0.33	0.39	0.22	**4.15**
	Niva	0.99	0.72	0.62	0.24	0.84	0.54	0.40	0.28	**4.63**
	Sparta	0.95	0.84	0.73	0.23	0.91	0.55	0.49	0.30	**5.00**
PR	Aries	0.87	0.64	0.47	0.63	0.83	0.38	0.34	0.26	**4.42**
	Maverick Gold	0.79	0.95	0.46	0.16	0.63	0.38	0.38	0.21	**3.96**
	Quartet	0.40	0.43	0.24	0.33	0.70	0.70	0.35	0.26	**3.41**
	Tonga	0.81	0.34	0.50	0.16	0.82	0.24	0.25	0.20	**3.32**
	Barfort	0.64	0.41	0.54	0.28	0.75	0.20	0.28	0.20	**3.30**
	Mara	0.77	0.58	0.50	0.23	0.74	0.45	0.37	0.53	**4.17**
	Calibra	0.60	0.56	0.43	0.63	0.65	0.47	0.25	0.22	**3.81**
TF	Bronson	0.69	0.70	0.90	0.74	0.86	0.65	0.78	0.47	**5.79**
	K5666V	0.79	0.52	0.83	0.42	0.58	0.29	0.30	0.22	**3.95**
	Barolex	0.82	1.02	1.03	0.46	0.68	0.38	0.54	0.30	**5.23**
	Mean	**0.8**	**0.6**	**0.6**	**0.4**	**0.8**	**0.4**	**0.4**	**0.3**	**4.3**
	CV	**46.2**	**38.9**	**39.0**	**92.3**	**48.9**	**42.3**	**35.6**	**36.1**	**18.4**
	LSD (0.05)	**0.6**	**0.4**	**0.4**	**0.5**	**0.6**	**0.3**	**0.2**	**0.2**	**1.3**

Table 12. Dry matter yield (tons acre^{-1}) of 16 grass cultivars established in monoculture at Lake City (LC) over a two-year period.

Grass Species	Cultivars	2004 19-May	2004 21-Jun	2004 5-Aug	2005 23-May	2005 7-Jul	2005 9-Aug	Total 2-yr
		DM tons acre^{-1}						
Fest	Duo	0.51	0.88	0.51	0.37	0.44	0.44	**3.15**
	Hykor	0.91	0.74	0.74	0.55	0.87	0.89	**4.70**
OR	Tekapo	0.73	0.69	0.85	0.18	0.57	0.27	**3.29**
	Amba	0.89	0.56	0.69	0.57	0.47	0.24	**3.42**
	Niva	0.71	0.70	0.77	0.46	0.26	0.34	**3.24**
	Sparta	1.12	0.71	0.86	0.61	0.52	0.51	**4.33**
PR	Aries	0.35	0.72	0.54	0.35	0.33	0.25	**2.54**
	Maverick Gold	0.14	0.45	0.62	0.69	0.35	0.42	**2.67**
	Quartet	0.42	0.84	0.43	0.33	0.34	0.43	**2.79**
	Tonga	0.50	0.44	0.32	0.44	0.47	0.32	**2.49**
	Barfort	0.28	1.25	0.61	0.20	0.48	0.29	**3.11**
	Mara	0.24	1.24	0.51	0.18	0.47	0.37	**3.01**
	Calibra	0.52	0.84	0.34	0.23	0.55	0.33	**2.81**
TF	Bronson	0.91	0.93	0.94	0.48	0.52	0.46	**4.24**
	K5666V	1.05	0.96	0.79	0.70	0.53	0.43	**4.46**
	Barolex	0.63	0.87	0.78	0.65	0.50	0.74	**4.17**
	Mean	**0.7**	**0.9**	**0.6**	**0.4**	**0.5**	**0.4**	**3.4**
	CV	**36.6**	**25.1**	**33.4**	**55.7**	**43.2**	**51.3**	**17.0**
	LSD(0.05)	**0.4**	**0.4**	**0.3**	**0.4**	**0.3**	**0.4**	**1.2**

Table 13. Dry matter yield (tons $acre^{-1}$) of 16 grass cultivars established in monoculture at Upper Peninsula Experiment Station (UPES) over a two-year period.

Grass Species	/ Cultivar	2004 7-Jun	2004 12-Jul	2004 22-Aug	2004 17-Oct	2005 1-Jun	2005 10-Jul	2005 23-Aug	2005 29-Sep	Total 2-yr
		DM tons $acre^{-1}$								
Fest	Duo	0.56	1.67	0.48	0.30	0.63	0.47	0.45	0.65	**5.21**
	Hykor	0.86	1.12	1.30	0.27	0.80	0.56	0.87	0.79	**6.57**
OR	Tekapo	0.94	1.33	0.96	0.31	0.72	0.64	0.60	0.77	**6.27**
	Amba	1.06	1.25	0.70	0.30	1.04	0.37	0.46	0.60	**5.78**
	Niva	0.78	1.09	0.93	0.28	0.97	0.66	0.70	0.57	**5.98**
	Sparta	0.82	1.11	1.01	0.30	1.01	0.56	0.62	0.54	**5.97**
PR	Aries	0.23	1.27	0.43	0.00	0.10	0.31	0.00	0.15	**2.49**
	Maverick Gold	0.36	0.78	0.58	0.00	0.00	0.37	0.00	0.28	**2.37**
	Quartet	0.42	1.76	0.35	0.00	0.16	0.33	0.12	0.42	**3.56**
	Tonga	0.59	1.44	0.74	0.00	0.60	0.42	0.48	0.54	**4.81**
	Barfort	0.75	1.93	0.59	0.00	0.67	0.30	0.54	0.77	**5.55**
	Mara	0.86	2.03	0.92	0.00	0.58	0.36	0.62	0.55	**5.92**
	Calibra	0.74	1.71	0.62	0.00	0.58	0.58	0.51	0.85	**5.59**
TF	Bronson	0.82	1.23	1.19	0.38	0.65	0.65	0.92	0.71	**6.55**
	K5666V	0.80	1.31	0.96	0.19	0.32	0.51	0.72	0.74	**5.55**
	Barolex	1.02	1.47	1.09	0.25	0.78	0.62	0.84	0.81	**6.88**
	Mean	**0.7**	**1.4**	**0.8**	**0.3**	**0.6**	**0.5**	**0.5**	**0.6**	**5.3**
	CV	**34.2**	**25.4**	**31.6**	**17.78**	**23.4**	**44.9**	**24.6**	**23.1**	**10.7**
	LSD(0.05)	**0.4**	**0.6**	**0.4**	**0.1**	**0.2**	**0.4**	**0.2**	**0.2**	**0.9**

Data of Perennial ryegrass cultivars in last cutting event of 2004 were not included

Table 14. Dry matter yield (tons $acre^{-1}$) of 67 grass- clover binary mixtures treatments established at Kellogg Biological Research (KBS) over a two-year period (continued on the next page).

		2004				2005				
Grass Cultivars	**Clover Cultivars**	24-May	24-Jun	26-Jul	9-Sep	4-May	14-Jun	2-Aug	13-Oct	**Total 2-yr**
		DM tons $acre^{-1}$								
Duo	Endura	0.91	0.53	0.52	0.72	0.48	0.55	0.28	0.28	**4.27**
Duo	Star Fire	0.57	0.67	0.77	0.63	0.75	0.71	0.30	0.24	**4.64**
Duo	VNS	0.65	0.70	0.62	0.71	0.85	0.65	0.30	0.38	**4.86**
Duo	Ladino	0.83	0.60	0.51	0.57	0.35	0.57	0.24	0.22	**3.89**
Duo	Kopu II	0.60	0.48	0.54	0.53	0.76	0.57	0.32	0.25	**4.05**
Hykor	VNS	0.92	0.84	0.66	0.61	0.99	0.65	0.68	0.43	**5.78**
Hykor	Jumbo	0.75	0.78	1.06	0.96	1.07	0.64	0.62	0.60	**6.48**
Hykor	Ladino	0.85	0.62	0.93	0.85	1.34	0.41	0.78	0.54	**6.32**
Amba	VNS	0.54	0.62	0.52	1.32	0.54	0.56	0.60	0.24	**4.94**
Amba	Jumbo	0.22	0.91	0.74	0.62	0.60	0.40	0.50	0.26	**4.25**
Amba	Ladino	0.66	0.61	0.75	0.79	0.74	0.40	0.54	0.34	**4.83**
Niva	VNS	0.51	0.64	0.66	0.82	0.60	0.48	0.55	0.36	**4.62**
Niva	Jumbo	0.75	0.64	0.66	0.77	0.50	0.45	0.55	0.30	**4.62**
Niva	Ladino	0.61	0.71	0.71	0.44	0.85	0.62	0.60	0.35	**4.89**
Sparta	VNS	0.73	0.75	1.02	0.70	0.83	0.48	0.50	0.36	**5.37**
Sparta	Jumbo	0.75	0.75	0.91	0.49	0.63	0.32	0.40	0.24	**4.49**
Sparta	Ladino	0.55	0.75	0.56	0.56	0.61	0.44	0.48	0.32	**4.27**
Tekapo	Endura	0.80	0.69	0.62	0.64	0.76	0.68	0.42	0.38	**4.99**
Tekapo	VNS	0.64	0.89	0.59	0.99	0.66	0.73	0.47	0.28	**5.25**
Tekapo	Star Fire	0.79	0.81	0.60	0.67	0.58	0.35	0.53	0.41	**4.74**
Tekapo	Kopu II	0.60	0.62	0.54	0.72	0.60	0.38	0.43	0.37	**4.26**
Tekapo	Ladino	0.94	0.60	0.73	0.57	0.73	0.70	0.45	0.34	**5.06**
Aries	Endura	0.53	0.61	0.44	0.37	0.59	0.40	0.30	0.32	**3.56**
Aries	VNS	0.62	0.48	0.47	0.65	0.48	0.26	0.43	0.26	**3.65**
Aries	Star Fire	0.78	0.44	0.83	0.92	0.53	0.55	0.38	0.30	**4.73**
Aries	Kopu II	0.62	0.61	0.40	0.52	0.65	0.51	0.40	0.28	**3.99**
Aries	Ladino	0.72	0.69	0.75	0.74	0.51	0.43	0.36	0.18	**4.38**
Barfort	VNS	0.99	0.56	0.69	0.55	0.40	0.37	0.35	0.34	**4.25**
Barfort	Start	0.64	0.58	0.56	0.68	0.55	0.96	0.35	0.22	**4.54**
Barfort	Alice	0.65	0.76	0.51	0.51	0.48	0.43	0.25	0.21	**3.80**
Barfort	Ladino	0.78	0.70	0.42	0.38	0.44	0.48	0.31	0.25	**3.76**
Calibra	VNS	0.54	0.44	0.46	0.98	0.78	0.37	0.32	0.36	**4.25**
Calibra	Jumbo	0.83	0.58	0.59	0.58	0.66	0.44	0.41	0.30	**4.39**
Calibra	Ladino	0.71	0.58	0.34	0.47	0.70	0.48	0.39	0.24	**3.91**
Mara	VNS	0.69	0.54	0.89	0.85	0.53	0.53	0.44	0.31	**4.78**
Mara	Start	1.05	0.73	0.32	0.65	0.65	0.37	0.51	0.33	**4.61**
Mara	Alice	0.74	0.66	0.57	0.62	0.43	0.50	0.43	0.40	**4.35**
Mara	Ladino	0.29	0.71	1.03	0.77	0.45	0.51	0.50	0.37	**4.63**
Mav. Gold	Endura	0.74	0.60	0.42	0.65	0.76	0.64	0.34	0.00	**4.15**
Mav. Gold	VNS	0.64	0.58	0.73	0.82	0.59	0.46	0.19	0.00	**4.01**
Mav. Gold	Star Fire	0.71	0.71	0.95	0.64	0.80	0.62	0.52	0.00	**4.95**
Mav. Gold	Kopu II	0.81	0.57	0.52	0.69	0.47	0.76	0.40	0.00	**4.22**
Mav. Gold	Ladino	0.65	0.66	0.44	0.61	0.48	0.75	0.39	0.00	**3.98**
Quartet	Endura	0.47	0.47	0.62	0.51	0.50	0.53	0.26	0.21	**3.57**
Quartet	VNS	0.99	0.40	0.63	0.50	0.52	0.38	0.30	0.36	**4.08**

Table 14 continued

		2004				2005				
Grass Cultivars	**Clover Cultivars**	24-May	24-Jun	26-Jul	9-Sep	4-May	14-Jun	2-Aug	13-Oct	**Total 2-yr**
Quartet	Ladino	0.78	0.61	0.53	0.65	0.50	0.55	0.28	0.22	**4.12**
Quartet	Star Fire	0.75	0.71	0.63	0.77	0.45	0.61	0.35	0.22	**4.49**
Quartet	Kopu II	0.89	0.60	0.44	0.59	0.24	0.33	0.25	0.25	**3.59**
Tonga	Endura	0.70	0.57	0.54	0.60	0.90	0.48	0.38	0.25	**4.42**
Tonga	VNS	0.52	0.60	0.69	0.89	0.78	0.50	0.31	0.20	**4.49**
Tonga	Star Fire	0.42	0.60	0.67	0.59	0.50	0.51	0.40	0.23	**3.92**
Tonga	Kopu II	0.60	0.65	0.84	0.64	0.43	0.45	0.33	0.18	**4.12**
Tonga	Ladino	0.59	0.60	0.67	0.59	0.58	0.34	0.26	0.12	**3.75**
Bronson	VNS	0.99	0.64	0.68	0.83	0.82	0.80	0.47	0.65	**5.88**
Bronson	Endura	0.73	0.59	0.87	0.95	1.20	0.84	0.51	0.60	**6.29**
Bronson	Kopu II	0.55	0.67	0.82	0.86	0.81	0.43	0.55	0.28	**4.97**
Bronson	Ladino	0.84	0.74	0.75	0.61	0.73	0.70	0.40	0.54	**5.31**
Bronson	Star Fire	0.64	0.67	0.44	0.86	1.03	0.64	0.55	0.58	**5.41**
K5666V	VNS	0.57	0.55	0.86	0.91	1.30	0.40	0.42	0.66	**5.67**
K5666V	Endura	0.97	0.72	0.55	0.54	0.64	0.52	0.27	0.40	**4.61**
K5666V	Kopu II	0.61	0.52	0.59	0.44	0.54	0.50	0.32	0.38	**3.90**
K5666V	Ladino	0.86	0.72	0.79	0.74	0.67	0.54	0.41	0.47	**5.20**
K5666V	Star Fire	0.59	0.54	0.65	0.88	0.71	0.68	0.36	0.47	**4.88**
Barolex	Alice	0.73	0.99	0.68	0.59	0.46	0.56	0.44	0.28	**4.73**
Barolex	VNS	0.73	0.77	0.84	0.69	0.64	0.75	0.69	0.51	**5.62**
Barolex	Ladino	0.51	0.85	1.07	0.79	0.74	0.76	0.61	0.40	**5.73**
Barolex	Start	0.93	0.70	0.99	0.85	0.69	0.74	0.51	0.56	**5.97**
	Mean	**0.7**	**0.6**	**0.7**	**0.7**	**0.6**	**0.5**	**0.4**	**0.3**	**4.6**
	CV	**33**	**30**	**37**	**41**	**36**	**36**	**26**	**41**	**16**
	LSD(0.05)	**0.4**	**0.3**	**0.4**	**0.4**	**0.4**	**0.3**	**0.2**	**0.2**	**1.2**

Grass cultivars:
Festulolium (Duo and Hykor)
Orchardgrass (Amba, Niva, Sparta and Tekapo)
Perennial ryegrass (Aries, Barfort, Calibra, Mara, Maverick Gold, Quartet, and Tonga,)
Tall fescue (Bronson, K5666V, and Barolex)

Clover cultivars:
Red clover (VNS*, Star Fire, and Start)
White clovers (KopuII, ladino, Alice, and Jumbo)
Kura clover (Endura)

* Variety Not Stated

Table 15. Dry matter yield (tons $acre^{-1}$) of 67 grass- clover binary mixtures treatments established at Lake City (LC) over a two -year period (continued on the next page).

		2004				2005				
Grass Cultivars	**Clover Cultivars**	19-May	21-Jun	5-Aug	7-Sep	23-May	7-Jul	9-Aug	2-Oct	**Total 2-yr**
		DM tons $acre^{-1}$								
Duo	Endura	0.64	0.99	0.27	0.41	0.23	0.63	0.29	0.28	**3.74**
Duo	Star Fire	1.00	0.81	0.36	0.71	0.46	0.38	0.35	0.31	**4.38**
Duo	VNS	0.74	1.00	0.67	0.83	0.41	0.38	0.48	0.34	**4.85**
Duo	Ladino	0.78	1.01	0.34	0.53	0.59	0.39	0.47	0.45	**4.56**
Duo	Kopu II	0.75	0.92	0.21	0.42	0.24	0.39	0.25	0.22	**3.40**
Hykor	VNS	1.08	0.71	0.59	0.76	0.66	0.23	0.33	0.54	**4.90**
Hykor	Jumbo	1.15	0.83	0.48	0.55	0.38	0.49	0.30	0.43	**4.61**
Hykor	Ladino	1.09	0.68	0.35	0.66	0.40	0.30	0.58	0.58	**4.64**
Amba	VNS	0.93	0.68	0.47	0.83	0.66	0.43	0.38	0.42	**4.80**
Amba	Jumbo	0.91	0.69	0.51	0.70	0.77	0.61	0.40	0.38	**4.97**
Amba	Ladino	0.83	0.67	0.36	0.78	0.72	0.63	0.37	0.34	**4.70**
Niva	VNS	1.00	0.58	0.51	0.91	0.56	0.37	0.33	0.49	**4.75**
Niva	Jumbo	0.72	0.70	0.35	0.85	0.58	0.54	0.22	0.45	**4.41**
Niva	Ladino	0.87	0.75	0.43	0.70	0.44	0.74	0.39	0.35	**4.67**
Sparta	VNS	0.83	0.69	0.54	0.65	0.40	0.44	0.26	0.38	**4.19**
Sparta	Jumbo	0.8	0.65	0.44	0.82	0.50	0.27	0.31	0.31	**4.10**
Sparta	Ladino	0.94	0.8	0.40	0.63	0.81	0.57	0.26	0.42	**4.83**
Tekapo	Endura	0.69	0.68	0.42	0.63	0.38	0.53	0.38	0.37	**4.08**
Tekapo	VNS	0.81	0.93	0.45	1.01	0.89	0.60	0.38	0.42	**5.49**
Tekapo	Star Fire	0.96	0.62	0.52	0.83	0.53	0.44	0.20	0.42	**4.52**
Tekapo	Kopu II	1.08	0.72	0.55	0.65	0.29	0.43	0.29	0.28	**4.29**
Tekapo	Ladino	0.78	0.62	0.59	0.54	0.46	0.65	0.21	0.22	**4.07**
Aries	Endura	0.65	0.68	0.21	0.00	0.56	0.36	0.25	0.43	**3.14**
Aries	VNS	0.54	0.97	0.97	0.00	0.79	0.48	0.23	0.34	**4.32**
Aries	Star Fire	0.81	0.96	0.48	0.00	0.51	0.40	0.21	0.34	**3.71**
Aries	Kopu II	0.47	0.92	0.49	0.00	0.26	0.33	0.22	0.45	**3.14**
Aries	Ladino	0.63	0.75	0.42	0.00	0.35	0.31	0.19	0.28	**2.93**
Barfort	VNS	0.83	1.14	0.55	0.00	0.42	0.36	0.25	0.32	**3.87**
Barfort	Start	0.75	1.05	0.51	0.00	0.23	0.31	0.35	0.27	**3.47**
Barfort	Alice	0.52	1.02	0.19	0.00	0.23	0.53	0.27	0.25	**3.01**
Barfort	Ladino	0.72	1.06	0.35	0.00	0.42	0.39	0.40	0.42	**3.76**
Calibra	VNS	0.59	1.40	0.68	0.00	1.07	0.92	0.30	0.40	**5.36**
Calibra	Jumbo	0.71	0.97	0.31	0.00	0.18	0.39	0.31	0.26	**3.13**
Calibra	Ladino	0.65	1.05	0.30	0.00	0.25	0.55	0.24	0.41	**3.45**
Mara	VNS	0.65	1.16	0.48	0.00	0.54	0.63	0.53	0.47	**4.46**
Mara	Start	0.54	1.25	0.48	0.00	0.44	0.39	0.49	0.46	**4.05**
Mara	Alice	0.62	1.22	0.42	0.00	0.23	0.40	0.41	0.61	**3.91**
Mara	Ladino	0.56	0.97	0.44	0.00	0.25	0.33	0.25	0.32	**3.12**
Mav. Gold	Endura	0.76	1.01	0.34	0.00	0.58	0.68	0.30	0.21	**3.88**
Mav. Gold	VNS	0.61	0.81	0.77	0.00	0.45	0.40	0.31	0.20	**3.55**
Mav. Gold	Star Fire	0.56	0.86	0.95	0.00	0.34	0.29	0.30	0.27	**3.57**
Mav. Gold	Kopu II	0.79	0.70	0.46	0.00	0.46	0.46	0.18	0.20	**3.25**
Mav. Gold	Ladino	0.45	1.00	0.47	0.00	0.70	0.42	0.14	0.26	**3.44**
Quartet	Endura	0.74	1.03	0.29	0.00	0.41	0.40	0.52	0.37	**3.76**
Quartet	VNS	0.75	0.89	0.79	0.00	0.36	0.37	0.49	0.46	**4.11**
Quartet	Ladino	0.69	1.01	0.55	0.00	0.58	0.35	0.35	0.14	**3.67**
Quartet	Star Fire	0.88	0.85	0.63	0.00	0.45	0.40	0.37	0.42	**4.00**

Table 15 continued

		2004				2005				
Grass Cultivars	**Clover Cultivars**	19-May	21-Jun	5-Aug	7-Sep	23-May	7-Jul	9-Aug	2-Oct	**Total 2-yr**
Quartet	Kopu II	0.68	1.11	0.42	0.00	0.51	0.56	0.41	0.35	**4.04**
Tonga	Endura	0.76	0.78	0.33	0.00	0.67	0.40	0.27	0.20	**3.41**
Tonga	VNS	0.83	0.62	0.70	0.00	0.29	0.80	0.37	0.35	**3.96**
Tonga	Star Fire	0.99	0.87	0.45	0.00	0.80	0.42	0.34	0.39	**4.26**
Tonga	Kopu II	0.74	0.67	0.27	0.00	0.78	0.32	0.26	0.29	**3.33**
Tonga	Ladino	0.62	0.79	0.35	0.00	0.78	0.43	0.32	0.66	**3.95**
Bronson	VNS	0.76	1.10	0.51	0.74	0.75	0.58	0.30	0.59	**5.33**
Bronson	Endura	0.79	0.72	0.45	0.80	0.39	0.53	0.48	0.46	**4.62**
Bronson	Kopu II	0.88	1.00	0.51	0.60	1.03	0.45	0.37	0.34	**5.18**
Bronson	Ladino	0.87	0.65	0.47	0.67	0.46	0.40	0.39	0.43	**4.34**
Bronson	Star Fire	0.78	0.82	0.55	1.11	0.69	0.32	0.39	0.43	**5.09**
K5666V	VNS	0.78	0.92	0.38	0.66	0.49	0.35	0.32	0.38	**4.28**
K5666V	Endura	0.89	0.86	0.48	0.47	0.55	0.39	0.46	0.50	**4.60**
K5666V	Kopu II	0.74	0.90	0.41	0.57	0.83	0.55	0.24	0.35	**4.59**
K5666V	Ladino	0.78	0.76	0.38	0.68	0.32	0.34	0.31	0.56	**4.13**
K5666V	Star Fire	0.76	0.73	0.55	0.59	0.34	0.29	0.28	0.42	**3.96**
Barolex	Alice	0.75	1.34	0.60	0.65	0.39	0.33	0.37	0.43	**4.86**
Barolex	VNS	0.86	1.16	0.80	0.76	0.74	0.37	0.20	0.39	**5.28**
Barolex	Ladino	0.66	0.97	0.49	0.80	1.03	0.22	0.40	0.49	**5.06**
Barolex	Start	0.61	0.69	0.54	0.97	0.74	0.37	0.24	0.34	**4.50**
	Mean	**0.8**	**0.9**	**0.5**	**0.7**	**0.5**	**0.4**	**0.3**	**0.4**	**4.5**
	CV	**30**	**28**	**42**	**33**	**53**	**43**	**52**	**37**	**14**
	LSD (0.05)	**0.4**	**0.4**	**0.3**	**0.4**	**NS**	**NS**	**NS**	**NS**	**NS**

Data of Perennial ryegrass cultivars in last cutting event of 2004 were not included

Grass cultivars:
Festulolium (Duo and Hykor)
Orchardgrass (Amba, Niva, Sparta and Tekapo)
Perennial ryegrass (Aries, Barfort, Calibra, Mara, Maverick Gold, Quartet, and Tonga,)
Tall fescue (Bronson, K5666V, and Barolex)

Clover cultivars:
Red clover (VNS*, Star Fire, and Start)
White clovers (KopuII, ladino, Alice, and Jumbo)
Kura clover (Endura)

* Variety Not Stated

Table 16. Dry matter yield (tons acre^{-1}) of 67 grass- clover binary mixtures treatments established at Upper Peninsula Experiment Station (UPES) over a two-year period (continued on the next page).

		2004				2005				
Grass Cultivars	**Clover Cultivars**	7-Jun	12-Jul	22-Aug	17-Oct	1-Jun	10-Jul	23-Aug	29-Sep	**2-yr Total**
		DM tons acre^{-1}								
Duo	Endura	0.37	0.31	0.64	0.23	0.58	0.35	0.34	0.31	**3.13**
Duo	Star Fire	0.79	0.72	0.81	0.40	0.91	0.64	0.83	0.30	**5.40**
Duo	VNS	0.69	0.72	0.37	0.28	0.70	0.50	0.50	0.37	**4.13**
Duo	Ladino	0.95	0.81	0.76	0.32	0.60	0.44	0.18	0.12	**4.18**
Duo	Kopu II	0.87	0.67	0.58	0.31	0.55	0.33	0.24	0.20	**3.75**
Hykor	VNS	0.71	0.47	0.43	0.29	0.74	0.33	0.52	0.38	**3.87**
Hykor	Jumbo	1.11	0.77	0.88	0.28	0.86	0.50	0.44	0.25	**5.09**
Hykor	Ladino	0.75	0.65	0.39	0.30	0.67	0.50	0.42	0.42	**4.10**
Amba	VNS	0.66	0.40	0.43	0.30	0.74	0.27	0.52	0.43	**3.75**
Amba	Jumbo	0.72	0.65	0.66	0.30	0.75	0.34	0.28	0.34	**4.04**
Amba	Ladino	0.77	0.35	0.44	0.31	0.77	0.20	0.28	0.37	**3.49**
Niva	VNS	0.68	0.49	0.63	0.27	0.70	0.37	0.38	0.40	**3.92**
Niva	Jumbo	0.64	0.71	0.57	0.29	0.56	0.40	0.27	0.27	**3.71**
Niva	Ladino	0.63	0.52	0.36	0.30	0.68	0.33	0.24	0.50	**3.56**
Sparta	VNS	0.76	0.48	0.51	0.30	0.57	0.24	0.34	0.35	**3.55**
Sparta	Jumbo	0.78	0.83	0.79	0.29	0.70	0.30	0.35	0.53	**4.57**
Sparta	Ladino	0.83	0.71	0.41	0.24	0.70	0.26	0.30	0.33	**3.78**
Tekapo	Endura	0.36	0.40	0.32	0.23	0.66	0.32	0.26	0.38	**2.93**
Tekapo	VNS	0.62	0.51	0.57	0.34	0.67	0.32	0.33	0.38	**3.74**
Tekapo	Star Fire	0.51	0.65	0.71	0.36	0.83	0.46	0.37	0.48	**4.37**
Tekapo	Kopu II	0.57	0.67	0.63	0.31	0.67	0.38	0.24	0.41	**3.88**
Tekapo	Ladino	0.60	0.73	0.59	0.26	0.66	0.30	0.25	0.41	**3.80**
Aries	Endura	0.31	0.42	0.77	0.00	0.50	0.40	0.24	0.11	**2.75**
Aries	VNS	0.51	0.46	0.83	0.00	0.51	0.60	0.60	0.13	**3.64**
Aries	Star Fire	0.35	0.76	0.79	0.00	0.73	0.44	0.40	0.31	**3.78**
Aries	Kopu II	0.55	0.62	0.46	0.00	0.55	0.34	0.30	0.10	**2.92**
Aries	Ladino	0.54	0.73	0.55	0.00	0.51	0.30	0.13	0.15	**2.91**
Barfort	VNS	0.64	0.74	0.51	0.00	0.62	0.70	0.64	0.36	**4.21**
Barfort	Start	1.26	0.74	0.55	0.00	0.62	0.66	0.53	0.28	**4.64**
Barfort	Alice	0.94	1.40	0.67	0.00	0.62	0.42	0.30	0.25	**4.60**
Barfort	Ladino	0.78	1.02	0.47	0.00	0.57	0.35	0.25	0.18	**3.62**
Calibra	VNS	0.58	0.55	0.51	0.00	0.61	0.26	0.44	0.30	**3.25**
Calibra	Jumbo	0.71	1.39	0.68	0.00	0.64	0.20	0.36	0.20	**4.18**
Calibra	Ladino	0.90	0.81	0.62	0.00	0.60	0.30	0.33	0.30	**3.86**
Mara	VNS	0.70	0.60	0.60	0.00	0.62	0.27	0.46	0.53	**3.78**
Mara	Start	0.74	0.86	0.80	0.00	0.70	0.64	0.50	0.52	**4.76**
Mara	Alice	0.72	1.16	0.72	0.00	0.62	0.31	0.21	0.25	**3.99**
Mara	Ladino	0.78	0.79	0.70	0.00	0.55	0.34	0.24	0.36	**3.76**
Mav. Gld	Endura	0.13	0.32	0.25	0.00	0.58	0.38	0.24	0.15	**2.05**
Mav. Gld	VNS	0.32	0.46	0.82	0.00	0.62	0.63	0.65	0.13	**3.63**
Mav. Gld	Star Fire	0.60	0.66	1.02	0.00	0.77	0.90	0.34	0.21	**4.50**
Mav. Gld	Kopu II	0.50	0.69	0.53	0.00	0.54	0.22	0.23	0.17	**2.88**
Mav. Gld	Ladino	0.52	0.86	0.61	0.00	0.55	0.15	0.17	0.12	**2.98**
Quartet	Endura	0.37	0.35	0.34	0.00	0.50	0.50	0.20	0.30	**2.56**
Quartet	VNS	0.58	0.73	0.56	0.00	0.65	0.55	0.38	0.32	**3.77**

Table 16 continued

		2004				2005				
Grass Cultivars	**Clover Cultivars**	7-Jun	12-Jul	22-Aug	17-Oct	1-Jun	10-Jul	23-Aug	29-Sep	**2-yr Total**
Quartet	Ladino	0.70	0.83	0.66	0.00	0.65	0.27	0.15	0.20	**3.46**
Quartet	Star Fire	0.65	0.88	1.02	0.00	0.82	0.50	0.52	0.40	**4.79**
Quartet	Kopu II	0.83	0.88	0.75	0.00	0.58	0.36	0.14	0.18	**3.72**
Tonga	Endura	0.40	0.44	0.37	0.00	0.68	0.46	0.30	0.33	**2.98**
Tonga	VNS	1.07	0.62	0.53	0.00	0.72	0.64	0.41	0.60	**4.59**
Tonga	Star Fire	0.88	0.73	0.84	0.00	0.80	0.54	0.70	0.34	**4.83**
Tonga	Kopu II	0.75	0.70	0.91	0.00	0.50	0.22	0.24	0.25	**3.57**
Tonga	Ladino	1.01	0.66	1.13	0.00	0.58	0.36	0.23	0.20	**4.17**
Bronson	VNS	0.75	0.38	0.52	0.25	0.60	0.47	0.62	0.43	**4.02**
Bronson	Endura	0.55	0.31	0.31	0.22	0.50	0.31	0.36	0.25	**2.81**
Bronson	Kopu II	0.83	0.67	0.55	0.29	0.63	0.42	0.38	0.32	**4.09**
Bronson	Ladino	0.77	0.64	0.57	0.44	0.68	0.22	0.40	0.30	**4.02**
Bronson	Star Fire	0.80	0.54	0.63	0.29	0.71	0.37	0.50	0.37	**4.21**
K5666V	VNS	0.55	0.45	0.54	0.28	0.70	0.41	0.45	0.46	**3.84**
K5666V	Endura	0.39	0.45	0.51	0.29	0.40	0.78	0.27	0.25	**3.34**
K5666V	Kopu II	0.63	0.54	0.70	0.32	0.55	0.33	0.36	0.26	**3.69**
K5666V	Ladino	0.60	0.83	0.80	0.16	0.58	0.40	0.35	0.40	**4.12**
K5666V	Star Fire	0.77	0.58	0.80	0.35	0.65	0.67	0.63	0.34	**4.79**
Barolex	Alice	0.69	0.58	0.58	0.32	0.70	0.58	0.47	0.50	**4.42**
Barolex	VNS	0.68	0.43	0.56	0.28	0.58	0.46	0.28	0.48	**3.75**
Barolex	Ladino	0.59	0.64	0.60	0.38	0.50	0.34	0.33	0.42	**3.80**
Barolex	Start	0.79	0.73	1.04	0.36	0.80	0.43	0.40	0.35	**4.90**
	Mean	**0.7**	**0.6**	**0.6**	**0.3**	**0.6**	**0.4**	**0.4**	**0.3**	**3.8**
	CV	**26**	**27**	**34**	**25**	**18**	**46**	**48**	**36**	**15**
	LSD(0.05)	**0.3**	**0.3**	**0.3**	**0.1**	**0.2**	**0.3**	**0.3**	**0.2**	**0.9**

Data of Perennial ryegrass cultivars in last cutting event of 2004 were not included

Grass cultivars:
Festulolium (Duo and Hykor)
Orchardgrass (Amba, Niva, Sparta and Tekapo)
Perennial ryegrass (Aries, Barfort, Calibra, Mara, Maverick Gold, Quartet, and Tonga,)
Tall fescue (Bronson, K5666V, and Barolex)

Clover cultivars:
Red clover (VNS*, Star Fire, and Start)
White clovers (KopuII, ladino, Alice, and Jumbo)
Kura clover (Endura)

* Variety Not Stated

Table 17. Dry matter yield (tons acre^{-1}) of eight clover cultivars established in monoculture at Kellogg Biological Research Station (KBS) over a two-year period.

Clovers Species / Cultivars		2004				2005			Total
		24-May	24-Jun	26-Jul	9-Sep	4-May	14-Jun	2-Aug	2-yr
		DM tons acre^{-1}							
Kura Clover	Endura	1.53	0.64	0.68	0.60	0.57	0.63	0.50	**5.15**
Red Clover	Star Fire	0.94	0.52	0.51	0.69	0.53	0.43	0.27	**3.89**
	Start	0.64	0.57	0.64	0.61	0.53	0.48	0.08	**3.55**
	VNS	0.65	0.50	0.70	0.70	0.56	0.42	0.16	**3.69**
White Clover	Kopu II	0.74	0.38	0.40	0.51	0.36	0.32	0.38	**3.09**
	Alice	0.56	0.60	0.63	0.45	0.42	0.37	0.36	**3.39**
	Jumbo	0.88	0.91	0.69	0.69	0.41	0.43	0.08	**4.09**
	Ladino	0.79	0.67	0.43	0.44	0.40	0.36	0.15	**3.24**
	Mean	**0.8**	**0.6**	**0.6**	**0.6**	**0.5**	**0.4**	**0.3**	**3.8**
	CV	**51.8**	**43.6**	**44.9**	**38.5**	**66.1**	**45.4**	**78.4**	**20.8**
	LSD (0.05)	**0.8**	**0.5**	**0.5**	**0.4**	**0.5**	**0.3**	**0.3**	**1.4**

Table 18. Dry matter yield production (tons acre^{-1}) of eight clover cultivars established in monoculture at Lake City (LC) over a two-year period.

Clovers Species/Cultivar		2004			2005				Total
		19-May	21-Jun	5-Aug	23-May	7-Jul	9-Aug	2-Oct	2-yr
		DM tons acre^{-1}							
*Kura Clover	Endura	0.57	0.90	0.25	0.38	0.82	0.21	0.11	**3.24**
Red Clover	Star Fire	0.80	0.58	0.70	0.00	0.00	0.00	0.00	**2.08**
	Start	0.71	0.68	0.80	0.00	0.00	0.00	0.00	**2.19**
	VNS	0.64	0.72	1.20	0.00	0.00	0.00	0.00	**2.56**
White Clover	Kopu II	0.59	0.88	0.59	0.00	0.00	0.00	0.00	**2.06**
	Alice	0.73	0.78	0.44	0.00	0.00	0.00	0.00	**1.95**
	Jumbo	0.73	0.79	0.55	0.00	0.00	0.00	0.00	**2.07**
	Ladino	0.63	0.84	0.49	0.00	0.00	0.00	0.00	**1.96**
	Mean	**0.7**	**0.8**	**0.6**	**0.0**	**0.0**	**0.0**	**0.0**	**0.0**
	CV	**22.2**	**29.6**	**27.4**	**0.0**	**0.0**	**0.0**	**0.0**	**0.0**
	LSD (0.05)	**0.3**	**0.4**	**0.3**	**0.0**	**0.0**	**0.0**	**0.0**	**0.0**

LSD is not available due to missing data

* Kura clover (Endura) is the only cultivar reported in 2005.

Table 19. Dry matter yield (tons acre^{-1}) of eight clover cultivars established in monoculture at Upper Peninsula Experiment Station (UPES) over a two-year period.

Clovers Species/Cultivar		**2004**			**2005**			**Total**
		7-Jun	12-Jul	22-Aug	1-Jun	10-Jul	23-Aug	**2-yr**
		DM tons acre^{-1}						
Kura clover	Endura	0.52	0.23	0.28	0.69	0.40	0.02	**2.14**
Red Clover	Star Fire	0.44	0.77	0.75	0.70	0.04	0.04	**2.74**
	Start	0.70	0.71	0.46	0.58	0.00	0.00	**2.45**
	VNS	0.29	0.50	0.68	0.38	0.05	0.00	**1.90**
White Clover	Kopu II	0.39	0.32	0.45	0.61	0.24	0.03	**2.04**
	Alice	0.40	0.32	0.55	0.53	0.25	0.08	**2.13**
	Jumbo	0.46	0.32	0.52	0.69	0.21	0.02	**2.22**
	Ladino	0.41	0.46	0.45	0.59	0.20	0.16	**2.27**
	Mean	**0.5**	**0.5**	**0.5**	**0.6**	**0.2**	**0.1**	**2.3**
	CV	**36.7**	**32.4**	**44.3**	**28.9**	**99.4**	**163.9**	**23.6**
	LSD (0.05)	**0.3**	**0.3**	**0.4**	**0.3**	**0.3**	**0.1**	**0.9**

Chapter 2

ANIMAL GRAZING PREFERENCE AND FORAGE QUALITY OF PERENNIAL COOL SEASON GRASSES AND CLOVERS GROWN IN MONOCULTURE AND BINARY MIXTURES UNDER ROTATIONAL GRAZING

ABSTRACT

Grass-legume forage pasture mixtures are important to animal production in the temperate United States. The objective of this study, which was conducted from 2004-06 at Hickory Corners, MI, was to assess animal grazing preferences and forage quality of introduced grass and clover cultivars grown in monoculture and binary mixtures. Results indicated that grazing preference depends on the type of pasture species and the species within the mixtures; binary mixtures were preferred over the monocultures. Higher crude protein (CP), lower acid detergent fiber (ADF), and neutral detergent fiber (NDF) were found in binary mixture treatments compared to grass and clover monoculture treatments. Perennial rye grass cultivars demonstrated higher forage quality and animal preferences when associated with clover cultivars. Tall fescue was less preferred by animals compared to perennial rye grass, particularly when grown in a monoculture. The results showed that animal preference of festulolium (Duo) monoculture was similar to that of perennial ryegrass cultivars. The binary mixtures of festulolium (Duo)-clover and perennial ryegrass-clover enhanced preferences and forage quality compared to monoculture festulolium and perennial ryegrass, which indicate these mixtures are a good choice for livestock producers to use for grazing. Total dry matter yield and animal preferences were not always positively correlated.

INTRODUCTION AND BACKGROUND

The most important forage legumes in the North-Central US are alfalfa (*Medicago sativa* L.), red clover (*Trifolium pratense* L.), birdsfoot trefoil (*Lotus corniculatus* L.), and white clover (*Trifolium repens* L.) (Knight, 1985). However, their ability to remain in the pasture under grazing is often limited (Forde et al., 1989; Van Keuren and Matches, 1988). Kura clover (*Trifolium ambiguim* Bieb.) is a perennial legume that has extensive rhizomes with a wide range of adaptation, which makes it potentially more suitable as a pasture crop and for soil conservation purposes (Bryant, 1974; Speer and Allison, 1985).

White clover (common and ladino types) is one of the most important legumes, which can be used for grazing. Beside its palatability and high forage quality, white clover also has the ability to tolerate continuous, heavy grazing (Spitaleri et al., 2003). This characteristic makes it successfully adapted for rotational grazing. White clover may be grazed continuously or rotationally and may be grazed closely (2.54 to 5.08 cm) (Penn State University Agronomy guide 2005-2006).

Plant performance in pastures cannot be fully understood without reference to animals. Animals tend to prefer and consume the most palatable plants first if given a choice from various forage species. Hence, animals are always selective in what they eat. The word "select" is defined as the action of choosing in preference to others (Cruz and Ganskopp, 1998).

Animal preferences refer to the selective response made by animals, which is mostly behavioral (Vallentine, 2001). Relative preference indicates a proportional choice among two or more feeds (Heady, 1964). Preference is important because it affects animal intake and any factor limiting intake can impact animal performance (McCaughey, 1998). On the other hand, consumption of the more palatable species can have a great impact on grass-legume binary mixture composition. Hence, animal preference is a factor that influences the pasture community as a result of overgrazing of the most preferred species in the pasture.

Animal grazing preference has been researched for many years. Several studies have investigated the factors that make animals prefer certain plant species over others. Some of these factors are plant-related while others are animal-related. Plant growth stage is a major factor that influences animal preference. Advanced growth stage has been indicated as a factor, which is correlated with decreasing preferences (Heady, 1964) Fresh immature forages is highly palatable and livestock will selectively graze those forages (Rohweder and Albrecht, 1995). The high preferences for rough fescue appeared to be determined by the accessibility of the large tufted plants to cattle (Willma and Rode, 1998). Animals graze the most palatable species when multiple species are offered. If no alternative is available, they will consume the specific feed even if it is less palatable.

Preference can be associated with plant physical characteristics. Within plant parts, young leaves and stems are higher in crude protein and lower in lignin than older stems and leaves (Heady, 1964). Thus, the younger leaves and stems are more likely to be preferred by animals. Animals avoid consuming plants with spines, pubescence or poisonous leaves. MacAdam and Mayland (2003) found that there was a negative correlation between tall fescue leaf strength and preference. They concluded that the cultivars that have the highest tensile strength would have the most fiber making it difficult to break down by grazing.

Plant chemical composition also influences animal preference. Forage species high in sugar content tend to be preferred by cattle (Plice, 1952), calves (Kare and Halpern, 1961), and deer (Mitchell and Hosley, 1936). Additionally, animals are able to preferentially select afternoon cut alfalfa hay over morning cut hay due to higher sugar content. Three ruminant species, sheep (*Ovis aries*), goats (*Capra hircus*), and cattle (*Bos taurus*) preferred alfalfa hay cut in the afternoon over hay cut in the morning (Fisher et al., 2002). High positive correlation between protein content and preference by cattle and sheep has been reported (Saltonstall, 1948; Cook, 1959; Blaser et al., 1960). In addition, lignin and crude fiber were observed to be negatively correlated with animal preference (Collins and Fritz, 2003). In most recent study, Smit et al., (2006) concluded that dairy cows selection among six perennial ryegrass cultivars was positively related to high water soluble carbohydrates (WSC) concentration and negatively to ash and fiber.

Past experience of animal diet selection influences animal preference. Vallentine (2001) stated in his book (Grazing Management) “Animals can acquire preferences for familiar food, first from their dams and second from their peers, but also by trial and error”.

Several methods have been reported to measure animal preferences. In general, the food that is eaten has been defined as the most preferable (Smit et al., 2006).

Preferences can be measured indoor by weighing the offered herbage and the residual then preferences is expressed as the food that has been consumed (offered- residual) (Aderibigbe et al., 1982; Provenza et al., 1996; Tolkamp et al., 1998). However, in a grazing system, preferences can be measured using the sward-cutting method (Meijs, 1981; Macoon., 2003). Animal preference can also be determined visually by estimating the herbage yield before and after grazing using preference scoring of 0 to 10 (0 to 100% of forage eaten) (Shewmaker et al., 1997) or a scale of 1 to 5 (1=least preferred ; 5 = most preferred) described by McCaughey (1998).

Forage quality is extremely important for livestock producers. The term forage quality is defined as the capacity of forage to supply animal nutrient requirements. Crude protein (CP), acid detergent fiber (ADF) and neutral detergent fiber (NDF) are the most common criteria that can be used to evaluate forage quality. Forage species with low NDF or ADF content are considered higher in quality than one with a high NDF or ADF content. In addition, as CP increases in forage, livestock perform greater (i.e., gain more weight, produce more milk, etc.) (Clemson, University Cooperative Extension Service). Buxton and Mertens (1995) defined forage quality in terms of performance of animals when fed herbage. It also refers to the physical and chemical characteristics of forage that make it valuable to animals as a source of nutrients (Balasko and Nelson, 2003). Collins and Fritz (2003) found that forage species vary in their quality components. They stated that "Legumes usually have 15% to 20% crude protein, while tropical forage grasses have about half as much and cool season grass had intermediate concentration ".

Differences in forage quality within grass and legume species may exist. Collins and Fritz (2003) reported that cool season grasses averaged about 13% higher in digestibility than warm season grasses. Leaf anatomy contributes to differences in forage quality. It has been found from leaf cross sections that bermudagrass (a C4 plant) is generally higher in the low digestible vascular bundle, epidermis and sclerechyma tissues than tall fescue (a C3 plant) (Akin and Burdick, 1975). Forage species can also differ in forage quality based on weather conditions. For instance, high temperatures decrease forage quality due to increased plant lignin content (Castle and Halley, 1953; Corbett, 1953). Anti-quality components are considered a major factor that decreases forage quality and animal preferences (Collins, and Hannaway, 2003).

Grass-legume mixtures vary in fiber depending on the species. Pastures consisting of kura clover with grasses had lower levels of fiber, greater protein concentration, and higher digestibility than the pasture consisting of red clover with grasses (Mouriño et al., 2003). It has been concluded that in a temperate pasture, white clover is selectively grazed in preference to perennial ryegrass (Curll and Wilkins, 1982). It was also observed that plant species selection was influenced by the relative maturities of the grass and legume (Grant.et al., 1985). Animals are able to select the legume from the lower quality grasses in a mixed stand of legume and grasses (Laidlaw, 1983).

Forage quality of the grass-legume binary mixtures appears to be higher than grass monocultures. Binary mixtures tend to have greater CP and lower NDF than pure stands of grass (Collins and Hannaway, 2003). In contrast, few studies reported that forage quality of grass-legume mixture is more likely to be lower in quality than the pure legume due to the addition of the grass component (Van Soest, 1982).

In recent years, both the grass and legume seed industry has began marketing new perennial grass and clover species and cultivars to Michigan growers. For example, orchardgrass has been introduced with value-added traits such as increased tillering, later maturity, and better winter hardness. Tall fescue has been recently offered with smoother leaf blades, endophyte free, and greater palatability. Because of the potential high forage dry matter yield of these species and cultivars, dairy and livestock producers may benefit by adapting these species and cultivars in their grazing systems.

Currently, there has been no research done to evaluate animal preferences and forage quality for introduced grass cultivars of festuloliums, orchardgrass, tall fescue and perennial ryegrass cultivars and clover cultivars of red clover, white clover and kura clover grown in a binary mixture. Therefore, more research is needed to determine animal preference and forage quality when grown in a mixture. Thus in this research, different grass and clover cultivars combinations were tested for their animal preference and forage quality under rotational grazing.

The objectives of this study were to determine animal preference of introduced grass and clover species and cultivars under rotational grazing, and determine forage quality based o NDF, ADF and CP, of the same grass and legume growing in monoculture and binary mixture under rotational grazing.

MATERIALS AND METHODS

Experiment Establishment and Maintenance:

The grazing trial was conducted in the Kellogg Biological Research Station at Hickory Corners, MI at the same experimental area established for evaluating persistence and productivity trial mentioned in Chapter 1.

The study area was separated into three portions: grass only, legume only (clover), and grass- clover binary mixtures (Appendix Fig. 2A). Each portion was arranged in a randomized complete block design (RCBD) with three replications. Each replication of the binary mixture portion consisted of 67 entries of different combinations of grass and clover mixtures. Each replication of clover and grass portion consisted of 8 and 16 monocultures of clover or grasses entries respectively (Chapter 1). Soil was analyzed at Michigan State University (MSU) soil laboratory for fertilizer recommendation as indicated in the first chapter. Appendix Table 1A presents soil information and the grass and clover cultivars used in this study.

Grazing procedure and data collection: All treatments were rotationally grazed during the 2004-2006 seasons. The grazing season began in late April and continued until early to late September, depending on the weather conditions. Grazing was started when the average grass height ranged from 20 to 30 cm. Four grazing events were typically obtained at the site of the study (Hickory Corners, MI). A total of eight grazing events were taken over the three years of the study. Four grazing events in 2004, three grazing events in 2005 and a single grazing event in 2006 were conducted.

For animal grazing, the whole area (binary mixture portion, grass and clover monoculture portions) was divided into two sections using electric fencing. One section consisted of replications 1 and 2 of the binary mixtures portion. The second section consisted of the replication 3 of the binary mixtures and all three replications of the monoculture portions (grasses and clovers). For all grazing events, animals began grazing in section 1 for 48 hr then they were moved to section 2 for another 48 hr. The grazing map of the arrangement used in this trial is illustrated in Appendix Fig 3A.

The number of cattle released to graze depended upon their body weight. In each year, approximately 10 Holstein steers, each weighting 226 kg was used for the first grazing event (Holstein heifers were used for 2006 grazing). However, for the subsequent grazing events, the number of animals was reduced to maintain the same stocking density (Appendix Table 5A). After each grazing event, plots were mechanically mowed to a uniform height (8 cm) and cattle were kept in different paddocks until the next grazing event. Adequate water and minerals were available during the grazing time in the grazing area.

Preference scores were visually assigned to assess the degree to which each of the plots had been grazed off after allowing the animals to graze each section for 48 hr. The preference rating was conducted by assessing the amount of remaining vegetation on a scale of (1-5) (McCaughey, 1998) where: **1** = less than 20% of the plot grazed; **2** = 20-40%; **3** = 40 - 60 %; **4** = 60 - 80 %; and **5** = 80% or greater of the plot was grazed. The cultivars that have high preference score over the course of the study are potentially more palatable.

Forage quality analyses: Prior to each grazing event, forage samples were hand clipped within 0.25 m^2 quadrate in each plot of the grass, clover and binary mixtures portions. Additional samples were also collected from Lake City Experiment Station (LC) and the Upper Peninsula Experiment Station at Chatham (UPES), MI.

Samples were dried at 60 °C for 48 hr for dry matter determination (Data on dry matter yield presented in Chapter 1). For forage quality analyses, dried samples were ground to pass through a 2 mm screen using a Wiley Grinding Mill (Authur H. Thomas Co., Philadelphia, PA) and then passed through a 1mm screen using a UDY Cyclone Mill, (Udy Mill Corp., Fort Collins, CO.).

In the crude protein analysis, total N was determined for the subset by the Hach modified Kjeldahl procedure (Watkins et al., 1987), and CP was estimated by multiplying total N by 6.25 because N makes up about 16% of the protein in the plant (Collins and Fritz, 2003). The Goering and Van Soest (1970) method was used for NDF and ADF determination with the addition of one ml of alpha-amylase to the neutral detergent solution for the breakdown of starch.

A sub-sample of ~20 grams of each sample was retained for forage quality analysis using Near-Infrared Reflectance Spectroscopy (NIRS). Each sample was scanned with a 6500 near-infrared spectrophotometer (FOSS NIRSystems, Inc., Eden Prairie, MN) with wavelengths between 400 and 2500 nm. Reflected wavelengths were recorded. CP, ADF and NDF were determined from equations developed by the NIRS consortium (Madison, WI). All the chemical analyses and NIRS procedures were performed at the MSU Forage and Physiology Lab.

Statistical analyses: All the statistical computations were performed using SAS (SAS Institute, 2000). Analysis of variance was conducted using PROC GLM procedure. Experimental units were the individual plots. The data for 2004 and 2005 were analyzed separately. Experiment type and grazing event were the fixed factors and blocks (replication) nested within type was a random factor used as an error term to test the effect of type. Grazing by blocks by type interaction term was used as an error term to test the effect of grazing and the effect of grazing by type interaction. When the interaction between grazing and type was found to be significant ($p<0.05$), mean separations between the types were conducted separately within each grazing.

Mean separation of animal preference scores and CP, ADF and NDF was achieved using Fischer's LSD at $p<0.05$. Normality assumption was checked using stem and leaf and normal probability plots in PROC UNIVARIATE. Coefficient of variation (CV) was listed to measure the precision of the experiment.

RESULTS AND DISCUSSION

Animal preferences

Significant years X grazing events X treatment (binary mixtures) interactions were found in animal preferences for grass-clover binary mixtures (Appendix Table 9A). Similar interactions were observed for grass monoculture (Appendix Table 10A). These interactions were primarily due to changes in plant maturity from one grazing event to the other and also from year to year. However, these interactions were not significant for clover monoculture (Appendix Table 11A).

In general, there was a trend toward higher preference scores of early grazing events compared to later grazing events in 2004 (Fig1) and 2005 (Fig2). During the early season, animals had less experience selecting among the forage species and as the season progressed, they started to acquire preferences for their diet selection (Vallentine, 2001). This may also be a result of lower plant lignin and/or higher sugar levels in the early spring.

Animals in this study showed varying degrees of preference between monocultures and binary mixtures. Results showed binary mixtures were preferred by the animals over monoculture treatments (Fig1 and 2). Binary mixtures were significantly higher in animal preference compared to grass monoculture ($p<0.01$) in three of four grazing events in 2004 and significantly ($p<0.05$) higher than clover monoculture in two grazing events in the same year (Fig 1). However, in 2005, grass-clover binary mixtures were significantly ($p<0.05$) higher than grass monoculture in one of three grazing events and higher than clover monoculture in two grazing events (Fig 2).

No significant differences were found between monoculture and binary mixtures in the single grazing event of 2006 (data not shown).

Animals showed a preference between grasses and clovers grown separately as monocultures. Clovers were significantly ($p<0.05$) greater in animal preference than the grasses in two grazing events in 2004 (Fig1). The grass monoculture was significantly preferred over the clover monoculture ($p<0.05$) in only the first grazing event in 2005 (Fig. 2). These data provide evidence of the importance of the binary mixtures as well as clovers for increased animal preference.

Grass monoculture data presented in Table 1 shows a significant difference in animal preference ($p<0.05$) between grass species and cultivars, while the preferences among clover species (Table 2) were less distinguishable than for grass species. Only one grazing event was significantly different in preferences rating among eight clover cultivars in 2004 (Table 2). In 2005, even though there were significant differences among the clovers cultivars across all the grazing events, the scores were lower for all grazing events compared to 2004 (Table 2), which is likely due to the higher maximum temperature causing increased lignification in grass and clover species. In addition, animal dry matter intake decreases with heat stress. In 2006, there was no data recorded on clovers due to the insufficient growth of the clover portion at the grazing event.

There was a significant difference in animal preference for the binary mixture treatments. The preference was more dependent upon the grass species than the associated clovers (Table 3). The animal preferences results of each grass species grown in monocultures or in binary mixtures are described as follows:

Festulolium: Duo grown in monoculture was significantly (p<0.05) higher in animal preference than Hykor in five of eight grazing events during the three year period (Table 1). When grown with clover in a binary mixture, Duo was significantly higher in animal preference (p<0.05) in six grazing events especially when associated with white clover (Kopu II) and kura clover (Endura) (Table 3). Duo was most likely preferred over Hykor because it has finer leaf blades, thus making it more desirable for animals (MacAdam and Mayland, 2003). In addition, since festuloliums are derived from a cross between Italian or perennial ryegrass and either meadow or tall fescue, it is likely that the genetic make up was a factor that influenced the difference in preference. Tall fescue has thicker and more course leaf blades than that of meadow fescue. Duo is the result of a cross of Meadow fescue and a tetraploid perennial ryegrass, with leaf blades and sugar content more like a ryegrass (AMPAC seed company web site). Thus, with the average of three grazing years, animal preference of Duo was similar to perennial ryegrass cultivars (Table1). These data confirm that Duo and Hykor festulolium cultivars differed in their animal preference and animals distinguish between them either when grown in monoculture or a binary mixture with a clover. However, even though Hykor appeared to be less palatable and had not been selected over Duo when they were grown in monocultures, it was grazed relatively well when grown in binary mixtures with clovers (Table 3). This would indicate that clovers in a binary mixture of somewhat unpalatable grasses can increase the consumption of the grasses which may not be normally preferred by animals due to the reduction of NDF in the binary as it associated with animal intake. There was no significant difference in animal preference between the three Hykor-clover binary mixtures (Table 3).

Orchardgrass: Hoveland, (1992) found that orchardgrass is a well suited perennial cool season grass for using in binary mixtures. Orchardgrass cultivars were less preferred by animals when grazed as a monoculture compared to binary mixtures of clover. Preference scores of four orchardgrass cultivars (Tekapo, Amba, Niva, and Sparta) were significantly ($p<0.05$) different in one of eight grazing events (Table 1). However, the binary mixture analysis presented in Table 3 showed greater preference for some cultivars in 2004 versus 2005 and 2006. For example, Tekapo had a very high preference score when grown with kura clover during the 2004 growing season and a low preference score for all the grazing events of 2005 and the single grazing event of 2006. As indicated earlier, the reduction of 2005 preference ratings may be due to weather related stress causing higher lignification and heat stress in animals. Orchardgrass cultivars Tekapo, Sparta, and Niva had lower preference scores when grown with certain white and red clover cultivars (Table 3). Over the three year grazing period, the best orchardgrass-clover combination was Tekapo with kura clover and with the white clover cultivar, Kopu II. Based on these data, Tekapo orchardgrass would be best suited as a binary mixture with clovers for Michigan producers under rotational grazing although Tekapo had lower persistence at the northern latitude whether grown in monoculture or binary mixtures (Chapter 1), which indicated that cultivar selection should be based on both the persistence and animal preference.

Perennial ryegrass: Perennial ryegrass is considered one of the most desirable species for grazing livestock because of its high palatability and forage quality (Balasko et al., 1995). Results showed that perennial ryegrass cultivars in the monoculture trial were high in animal preference in the first grazing event of 2004 and 2005 grazing seasons

(Table1). There was a significant difference in animal preference (p<0.05) among the seven perennial ryegrass cultivars when grown in monoculture (Aries, Maverick Gold, Quartet, Tonga, Barfort, Mara, and Calibra) at only two of eight grazing events (Table 1). Animals showed higher (p<0.05) selectivity for perennial rye grass cultivars in all grazing events over the three years period when grown in binary mixtures with clovers. For example, Aries-kura (Endura), Quartet-white clover (Kopu II), Quartet-kura (Endura) and Tonga -white clover (Ladino) binary mixtures resulted in the highest preference among perennial ryegrass-clover mixtures (Table 3)

Perennial ryegrass does not tolerate heat stress or drought and its production during hotter and drier summer months will be lower (Balasko and Nelson, 2003). This explains the decline in average preference during the second, third, and fourth grazing events of 2004 (Table 1). The preference of forages varies with seasonal conditions and stage of growth (Am.Soc.Range Mgmt, 1962).

Tall fescue: Tall fescue is considered less preferred by animals compared to perennial rye grass particularly when grown in monoculture. There was one grazing event which resulted in a significant difference (p<0.05) among the three monoculture tall fescue cultivars in this study (Bronson, K5666V, and Barolex) (Table 1). Tall fescue cultivars resulted in higher preference scores when grown in binary mixtures. There were significant differences between the tall-fescue binary mixtures (Table 3). Bronson and Barolex had significantly lower animal preferences when grown with red clover. Kura clover grown with K5666V resulted in the highest preference among the tall fescue mixtures.

Tall fescue cultivars demonstrated greater yield production, better compatibility with clovers, better winter hardness and good ground cover compared to other grass species in this study (Chapter 1), yet they were less preferred by animals compared to the other grass species even when grown in binary mixtures. The reason for the decreased preference for tall fescue cultivars was likely due to increased leaf width and thickness as explained by MacAdam and Mayland (2003). However, if animals are only offered tall fescue cultivars in pastures, they will likely consume it. Some of the cultivars in this experiment displayed leaf blades, which were narrower and less thick than others. However, it would be still more useful for Michigan growers to use tall fescue cultivars with clovers to enhance preference.

Forage Quality

Significant years X location X cutting (grazing) X treatment (binary mixtures) interactions were present for CP, ADF and NDF (Appendix Table 9A). These interactions were due primarily to change in sward composition of the mixtures, which occurred from one grazing event to another and from year to year, which was similar to results found by Zemenchik et al. (2001). Significant years X location X cutting (grazing) X treatment interactions were found for CP, ADF and NDF for grass and clover monoculture treatments (Appendix Tables 10A and 11A, respectively), which was due to the change in plant forage quality that occurred between grazing events and year to year. However, there was no interaction for (NDF) in grass treatments (Appendix Table 10A) and in (CP) clover (Appendix Table 11A). Results from forage quality analysis have proven the hypothesis, which states that binary mixtures have higher forage quality than monocultures (grass or clover). Results of forage quality show that clovers had highest

CP, lowest NDF and ADF compared to grass monocultures and binary mixtures (Table 4). Grass monoculture had lower forage quality (Greater ADF and NDF and lower CP) than binary mixtures. Combining clover with any of the cool-season grasses significantly reduced concentration of NDF and ADF and increased CP compared to grass monocultures (Table 5). These results concur with Zemenchick et al., (2002) in which they found that legume proportions in binary mixtures were positively correlated to CP concentrations and negatively correlated to NDF concentrations.

There was a significant difference in forage quality among species as well as between cultivars of each grass. In 2003, the first grazing event resulted in low ADF and NDF content in perennial ryegrass (Barfort) and orchardgrass (Tekapo) and higher CP than other grass cultivars when grown in binary mixtures. Warnock (2004) concluded that Barfort perennial ryegrass and Tekapo orchardgrass had lower NDF and ADF values than other cultivars such as perennial ryegrass (Aries). The 2004-2005 analysis of data revealed higher forage quality in all perennial ryegrass cultivars especially Maverick Gold, Quartet and Tonga. The forage quality of festulolium (Duo) was higher in CP and lower in ADF and NDF than that of Hykor festulolium. In addition, tall fescue (Bronson) and festulolium (Hykor) cultivars were lower in forge quality compared to the other cultivars within their species (Table 5). In the orchard grass monocultures, Sparta had the highest CP compared to Amba, Niva and Tekapo.

Clover species also were significantly different in forage quality. Kura clover (Endura) and white clover (Kopu II, Alice, Jumbo and Ladino) were higher in CP and lower in ADF and NDF compared to all red clover cultivars (Table 6). However, all red clover cultivars were much higher in CP and lower in ADF and NDF compared to perennial grass species.

Total yield and animal preferences were not positively correlated. For example, white clover (Jumbo)- festulolium (Hykor) mixtures that produced the highest total dry matter yield were less preferred by animals than perennial ryegrass-clover mixtures, which had lowest yield and a higher animal preferences rating (Data not shown). These results concur with a study conducted by Shewmaker et al. (1997).

CONCLUSIONS

Livestock producers should consider adapting forage species and cultivars, which show higher animal preferences and have higher forage quality for profitable systems. Thus, cultivar selection should be based upon animal preference, persistence and yield. The binary mixtures of festulolium (Duo)-clover increased preferences and forage quality compared to monoculture festulolium. In addition, perennial ryegrass cultivars demonstrated good forage quality and animal preferences when associated with clover cultivars, which indicate these mixtures are a good choice for livestock producers to use for grazing. However, their ability to survive the harsh winter could be an issue especially, when grow in northern latitudes (44^{o}-46^{o} N). Cultivars that have shown a higher preference to animals with higher forage quality (Aries, Maverick Gold) were more vulnerable to the winter injury. Therefore, areas where winter injury is problem, these cultivars would not be a good choice (Chapter 1).

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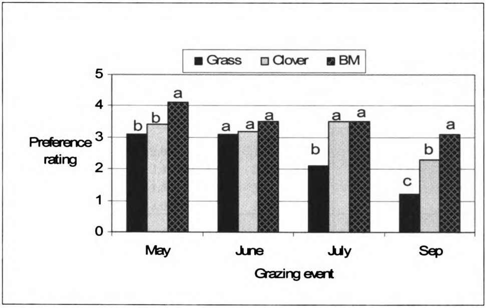

Fig.1. The differences in animal grazing preference for the binary mixture (BM), clover, and grass treatments during 2004 grazing season (Bars with different letters within grazing event are significantly different at $p<0.05$).

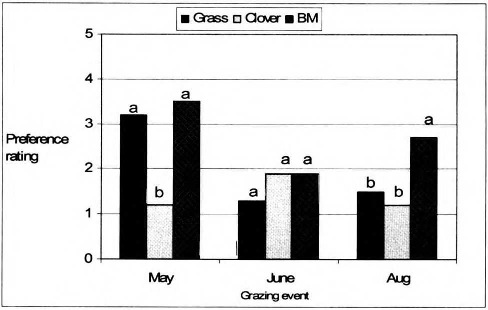

Fig 2. The differences in animal grazing preference for the binary mixture (BM), clover, and grass treatments during 2005 grazing season (Bars with different letters within grazing event are significantly different at $p<0.05$).

Table 1. Animal preference score of 16 grass cultivars established in monoculture treatments over eight grazing events.

Grass Species	Grass Cultivars	2004 28-May	2004 26-Jun	2004 28-Jul	2004 9-Sep	2005 6-May	2005 16-Jun	2005 4-Aug	2006 12-May
Fest	Duo	4.8	3.3	1.2	1.1	4.7	1.0	2.7	3.0
	Hykor	1.5	2.1	1.7	1.1	1.7	1.7	1.0	1.2
OR	Tekapo	2.0	1.5	1.3	1.0	1.5	1.0	1.2	2.3
	Amba	2.3	3.5	2.6	1.0	2.7	1.0	1.0	1.7
	Niva	1.5	3.5	3.3	1.8	2.0	1.0	1.0	3.2
	Sparta	3.0	3.0	3.3	1.8	1.9	1.3	1.0	3.2
PR	Aries	3.8	3.6	1.5	1.0	4.5	1.5	1.7	1.8
	Maverick Gold	4.8	3.5	2.0	1.5	4.2	1.0	2.2	1.2
	Quartet	4.0	3.0	1.8	1.0	3.5	1.0	2.0	1.5
	Tonga	4.0	3.5	1.5	1.0	4.8	2.0	2.4	3.3
	Barfort	4.8	3.6	1.5	1.3	4.7	1.3	1.7	3.3
	Mara	5.0	3.5	1.6	1.1	4.4	1.0	1.0	3.0
	Calibra	4.1	3.1	1.6	1.0	4.7	1.0	2.4	3.3
TF	Bronson	1.3	2.6	3.0	1.0	1.2	1.3	1.0	1.0
	K5666V	1.3	2.6	3.3	1.0	2.4	1.5	1.0	1.2
	Barolex	1.0	2.8	2.0	1.0	2.4	1.0	1.0	1.3
	Mean	**3.1**	**3.1**	**2.1**	**1.2**	**3.2**	**1.3**	**1.5**	**2.2**
	CV	**21**	**30**	**38**	**34**	**20**	**54**	**42**	**47**
	LSD(0.05)	**1.1**	**1.6**	**1.3**	**0.7**	**1.0**	**NS**	**1.0**	**1.7**

1 = less than 20% of the plot grazed

5 = 80% or greater of the plot was grazed.

Grass cultivars:

Fest=Festulolium ; OR=Orchardgrass, PR=Perennial ryegrass and TF= Tall fescue

Table 2. Animal preference score of eight clover cultivars established in monoculture over seven grazing events.

Clover Species	Clover Cultivars	2004 28-May	2004 26-Jun	2004 28-Jul	2004 9- Sep	2005 6-May	2005 16-Jun	2005 4-Aug
Kura	Endura	3.8	3.0	3.3	1.3	1.0	3.7	2.5
Red Clover	Star Fire	3.6	4.1	4.1	1.8	1.0	1.4	1.0
	Start	2.8	3.0	3.6	1.6	1.2	1.5	1.0
	VNS	2.6	2.8	2.5	2.5	1.0	1.7	1.0
White Clover	Kopu II	3.8	3.6	3.8	3.3	1.7	1.9	1.2
	Alice	3.8	3.0	3.5	2.5	1.0	1.0	1.0
	Jumbo	3.0	2.3	3.6	2.1	1.7	3.2	1.0
	Ladino	3.3	4.3	3.8	2.8	1.2	1.0	1.0
	Mean	**3.3**	**3.2**	**3.5**	**2.2**	**1.2**	**1.9**	**1.2**
	CV	**37**	**48**	**37**	**41**	**32**	**57**	**18**
	LSD (0.05)	**NS**	**NS**	**NS**	**1.6**	**0.7**	**1.9**	**0.4**

1 = less than 20% of the plot grazed
5 = 80% or greater of the plot was grazed.

Table 3. Animal preference score of 67 grass-clover binary mixtures over eight grazing events (continue next page).

Grass Cultivars	Clover Cultivars	2004				2005			2006
		28-May	26-Jun	28-Jul	9-Sep	6-May	16-Jun	4-Aug	12-May
Duo	Endura	4.8	3.3	3.0	2.8	3.2	2.5	4.8	5.0
Duo	Star Fire	4.5	3.3	2.5	2.8	4.4	1.0	2.5	1.3
Duo	VNS	4.8	3.1	2.3	2.1	3.0	1.2	1.9	2.7
Duo	Ladino	4.6	2.8	2.8	3.1	4.5	1.0	3.7	2.7
Duo	Kopu ll	4.8	4.6	3.1	4.0	4.7	1.0	2.0	3.0
Hykor	VNS	2.3	2.3	2.3	1.6	3.6	1.5	1.0	1.0
Hykor	Jumbo	2.3	2.3	3.5	2.0	3.3	1.0	1.0	1.3
Hykor	Ladino	2.6	2.3	3.3	1.8	3.5	1.3	1.5	1.3
Amba	VNS	3.8	3.8	3.8	3.0	3.3	3.0	1.3	1.0
Amba	Jumbo	3.8	4.5	4.1	4.1	3.8	1.8	2.2	1.0
Amba	Ladino	4.6	4.6	4.1	3.5	4.1	1.7	1.7	1.0
Niva	VNS	3.5	3.5	3.8	3.0	2.0	2.0	1.5	2.3
Niva	Jumbo	4.3	3.6	3.5	3.6	1.4	1.7	1.4	2.5
Niva	Ladino	3.3	4.3	4.3	3.3	2.7	2.0	2.0	2.7
Sparta	VNS	3.8	3.8	3.5	3.6	2.7	1.0	1.0	2.0
Sparta	Jumbo	3.8	4.5	4.5	4.0	3.4	3.0	1.9	3.2
Sparta	Ladino	3.8	2.8	4.5	3.3	2.9	1.5	1.9	2.2
Tekapo	Endura	3.8	4.0	5.0	4.5	2.5	3.9	3.5	2.2
Tekapo	VNS	4.0	2.8	3.6	2.8	1.4	1.7	1.4	2.7
Tekapo	Star Fire	3.8	2.1	2.0	3.5	2.0	3.4	2.2	1.2
Tekapo	Kopu ll	4.1	4.5	4.5	3.6	2.4	3.0	3.5	2.0
Tekapo	Ladino	4.6	3.1	4.8	3.8	1.7	2.5	2.2	1.2
Aries	Endura	4.6	4.5	4.0	4.8	4.2	3.7	4.5	3.3
Aries	VNS	4.1	3.8	3.5	4.0	3.2	2.8	3.0	2.2
Aries	Star Fire	3.8	3.8	3.8	3.0	4.4	2.2	3.7	2.0
Aries	Kopu ll	3.6	4.6	4.5	4.8	3.4	1.8	3.7	1.2
Aries	Ladino	4.8	3.5	3.1	3.0	4.2	1.5	3.0	2.3
Barfort	VNS	4.5	3.1	1.3	2.3	3.8	1.8	3.0	2.0
Barfort	Start	4.5	3.6	4.1	3.0	3.83	1.0	3.7	1.7
Barfort	Alice	4.0	3.8	3.6	3.8	3.7	1.2	3.7	2.3
Barfort	Ladino	4.6	4.3	4.1	3.1	4.0	1.5	2.5	2.5
Calibra	VNS	4.8	3.3	2.1	3.0	4.4	1.5	1.9	1.8
Calibra	Jumbo	4.3	4.3	3.3	3.8	5.0	1.4	4.0	2.0
Calibra	Ladino	5.0	4.1	4.1	2.6	2.7	2.7	4.7	2.7
Mara	VNS	4.0	2.0	1.5	1.5	3.5	1.5	2.5	4.3
Mara	Start	4.3	2.6	1.8	2.6	4.5	1.0	1.4	2.5
Mara	Alice	5.0	3.1	2.1	2.6	4.5	1.0	2.9	3.3
Mara	Ladino	4.5	2.1	2.0	2.3	4.5	1.0	2.4	3.5
Mavrick Gold	Endura	4.8	3.8	3.6	4.0	3.7	2.4	2.7	2.2
Mavrick Gold	VNS	4.1	2.8	2.1	2.3	3.0	2	2.9	1.5
Mavrick Gold	Star Fire	4.1	3.3	3.1	2.6	3.7	1.2	2.4	1.0
Mavrick Gold	Kopu ll	4.5	4.0	3.8	4.5	4.4	1.0	2.7	2.2
Mavrick Gold	Ladino	4.3	4.0	2.6	3.8	4.0	1.9	3.0	1.5
Quartet	Endura	4.8	4.0	4.3	4.5	4.5	3.9	4.7	2.5

Table 3 continued

Grass Cultivars	Clover Cultivars	2004				2005			2006
		28-May	26-Jun	28-Jul	9-Sep	6-May	16-Jun	4-Aug	12-May
Quartet	VNS	4.6	4.5	4.1	4.5	2.9	2.2	3.2	2.0
Quartet	Ladino	4.6	4.6	3.5	3.6	3.5	1.0	3.9	1.5
Quartet	Star Fire	4.8	4.6	4.6	3.3	3.5	1.4	3.5	1.5
Quartet	Kopu ll	5.0	4.5	4.8	2.6	3.4	3.7	4.5	2.0
Tonga	Endura	4.6	3.6	2.6	2.5	3.5	1.9	4.2	3.2
Tonga	VNS	4.0	3.5	2.8	1.8	4.0	1.0	4.0	2.0
Tonga	Star Fire	4.3	4.5	3.5	3.3	3.5	2.9	3.2	1.8
Tonga	Kopu ll	4.5	3.8	3.6	3.1	4.5	1.6	2.7	2.5
Tonga	Ladino	4.8	4.6	4.6	3.3	4.0	3.2	4.7	2.3
Bronson	VNS	2.5	1.6	3.0	1.1	3.4	1.7	1.0	1.0
Bronson	Endura	2.8	2.1	3.1	1.8	3.7	3.5	2.7	1.0
Bronson	Kopu ll	2.6	2.1	4.0	2.0	3.2	1.5	1.4	1.0
Bronson	Ladino	3.6	2.5	4.1	3.1	4.0	2.9	1.4	1.0
Bronson	Star Fire	3.5	2.0	3.1	1.6	2.9	1.0	1.0	1.3
K5666V	VNS	3.6	3.3	4.5	2.6	3.7	1.0	1.7	1.7
K5666V	Endura	4.0	3.8	4.6	4.0	4.2	2.5	4.7	2.3
K5666V	Kopu ll	3.8	4.1	5.0	4.5	3.9	1.3	2.9	1.8
K5666V	Ladino	4.1	3.1	3.8	2.1	3.7	2.4	2.7	1.3
K5666V	Star Fire	3.3	2.5	3.5	2.6	3.4	1.7	3.2	1.0
Barolex	Alice	3.3	4.0	3.8	3.1	4.8	1.5	2.2	1.7
Barolex	VNS	2.5	3.5	2.6	1.6	4.2	1.8	1.5	1.2
Barolex	Ladino	3.5	3.5	3.1	1.6	3.7	1.2	1.0	1.0
Barolex	Start	2.5	2.6	3.0	1.5	2.5	1.2	1.0	1.3
	Mean	**4.1**	**3.5**	**3.5**	**3.1**	**3.5**	**1.9**	**2.6**	**1.9**
	CV	**18.1**	**26.1**	**30.5**	**33.3**	**26.6**	**53.5**	**37.3**	**51.7**
	LSD (0.05)	**1.2**	**1.5**	**1.7**	**1.6**	**1.5**	**1.6**	**1.6**	**1.7**

1 = less than 20% of the plot grazed
5 = 80% or greater of the plot was grazed

Grass cultivars:
Festulolium (Duo and Hykor)
Orchardgrass (Amba, Niva, Sparta and Tekapo)
Perennial ryegrass (Aries, Barfort, Calibra, Mara, Maverick Gold, Quartet, and Tonga,)
Tall fescue (Bronson, K5666V, and Barolex)

Clover cultivars:
Red clover (Start Fire, Start and VNS)
White clover (Ladino, Alice, Kopu II, and Jumbo)
Kura (Endura)

Table 4 . The mean values (%) of forage quality components (CP, ADF and NDF) for pasture species of grass, clover and binary mixtures over 2004-2005 growing seasons in three different locations.

Pasture Type	2004			2005		
	CP [x]	ADF [y]	NDF [z]	CP	ADF	NDF
Grass	16.9	28.2	45.6	20.2	28. 2	54.2
Clover	20.2	26.4	33.4	20.6	20.9	29.8
Binary Mixtures	20.1	25.9	43.9	17.7	25.8	42.4

Values based on the (Lsmeans) of all locations and cuts
x Crude protein
y Acid detergent fiber (ADF)
z Neutral detergent fiber (NDF)

Table 5. Average forage quality parameters (CP, ADF and NDF) of 16 grass cultivars established in monoculture and binary mixtures in three locations.

Grass Species	Grass Cultivars	Monoculture			Binary Mixtures		
		CP	ADF	NDF	CP	ADF	NDF
				%			
Fest	Duo	19.4	25.6	48.2	23.6	22.1	37.1
	Hykor	17.6	29.7	51.5	20.5	25.2	44.6
OR	Tekapo	19.2	27.8	52.2	22.5	24.3	42.7
	Amba	18.9	26.1	49.3	21.7	23 .7	41.9
	Niva	19.3	25.2	50.7	22.4	24.3	42.9
	Sparta	20.2	26.4	50.2	22.3	23.8	42.4
PR	Aries	19.1	24.9	48.8	23.7	22.2	36.7
	Maverick Gold	20.6	23.9	49.1	23.5	22.0	373
	Quartet	20.1	25.1	45.7	24.3	21.4	35.7
	Tonga	20.1	23.4	46.2	24.0	21.5	35.5
	Barfort	19.8	23.7	46.9	23.7	21.7	36.4
	Mara	18.8	25.8	49.5	22.3	23.2	39.7
	Calibra	19.4	25.8	47.4	23.6	22.0	36.9
TF	Bronson	18.6	29.3	54.3	21.3	24.5	43.8
	K5666V	18.5	28.1	50.3	22.4	23.2	40.2
	Barolex	17.5	28.9	52.8	21.6	24.1	42.5

Fest= Festuloliums.
OR= Orchardgrass.
PR=Perennial ryegrass.
TF= Tall fescue.

Table 6. Average forage quality parameters (CP, ADF and NDF) of eight clover cultivars established in monoculture in three locations.

Clover		**Forage Quality Parameters**		
Species	**Cultivars**	**CP**	**ADF**	**NDF**
		%		
Kura	Endura	24.3	21.7	27.9
Red Clover	Star Fire	23.7	23.1	29.8
	Start	21.6	21.9	28.5
	VNS	23.9	22.9	29.3
White Clover	Kopu II	24.5	20.6	26.0
	Alice	25.3	20.3	26.1
	Jumbo	25.3	20.2	25.1
	Ladino	25.0	20.1	26.1

Chapter 3

PREDICTING BOTANICAL COMPOSITION OF GRASS-CLOVER PASTURES MIXTURES USING NEAR-INFRARED REFLECTANCE SPECTROSCOPY

ABSTRACT

Hand separation is a common method used to determine the botanical species composition of grass-legume swards. However, this method is very laborious and is time consuming. This study was conducted to evaluate the use of Near-Infrared Reflectance Spectroscopy (NIRS) to predict the species composition of several grass-clover binary mixtures with samples collected from three locations and over three-year period. Samples were taken during 2003-2005 growing season from clover-grass binary mixture experiments at Kellogg Biological Research Station at Hickory Corners (KBS), Lake City Research Station (LC) and Upper Peninsula Experiment Station at Chatham (UPES), MI. Second cutting event from each location was hand separated into grass, clover and weeds. All samples were dried, ground and scanned with NIRS. Three calibration equations were developed to predict the species composition. The first equation was developed using artificially mixed samples in 10% increments of pure grass and clover samples collected from monoculture plots at LC location in 2004. The second equation was created from hand-separated samples clipped at LC location in 2004. The third equation was created from selected subsets of hand-separated samples collected from all three locations and three years. The equations developed based on artificially mixed samples and the hand-separated samples from the single location and year resulted in a poor prediction of grass and clover components. Prediction coefficients of determination (R^2) of grass and clover were, 0.24 and 0.31, respectively for artificially mixed samples

equation and were equal 0.25 and 0.37 for the hand-separation equation. Standard errors of prediction (SEP) were relatively high for both calibration equations. However, the equations developed based on selected hand-separated samples from all three locations and all three years had higher prediction accuracy with R^2 ranging from 0.67 to 0.72 and SEP from 6.9 to 12.8, respectively. These results suggest that using either artificially mixed or hand separated samples from a single location in a single year is unsuitable to predict species composition at different locations and years. It can be concluded that NIRS can be applied to replace the hand separation method using the calibration equation developed from hand separation data from different locations and years with some limitations.

INTRODUCTION AND BACKGROUND

Legume content in a binary mixture is a key parameter for the quantification of N_2 fixation and diet quality. Pastures with less than 30% legume should either be fertilized with up to 168 kg of N ha^{-1} or improved by introducing legumes in the sward. Determination of botanical composition is important, since animal performance is dependent on the proportion of desired plant species in the diet (Petersen et al., 1987).

The development of a reliable and rapid approach for determining botanical composition has been a research goal for many years. There are several indirect methods for estimating botanical composition of grass and legume mixtures including visual estimates (Marten, 1964; Tanner et al., 1966; Tiwari et al., 1963), point quadrate methods (Leasure, 1949; Vankeuren and Ahlgren 1957), dry-weight rank method (Mannetij and Haydock, 1963; Walker, 1970) and chemical composition based method (Cooper et al., 1957). The method that is commonly used for determining botanical composition is hand-separation (Vankeuren and Ahlgren, 1957). Hand separation requires manually separating the mixture into its components such as legumes, grasses, and weeds and the component percentage is then determined by dry matter weight. However, hand-separation is not practical since it is laborious, time-consuming, costly, and prone to operator errors, especially when a large number of samples are being processed. Up to 2 to 3 hr may be required for identification and separation of sown pasture samples (Grant, 1981). Therefore, Near Infrared Reflectance Spectroscopy (NIRS) may be able to replace hand-separation as a rapid and more convenient technique.

The primary use of NIRS in forages is to determine nutrient composition values of feedstuffs (Norris et al., 1976). NIRS is an inexpensive method, which allows the dried forage samples to be stored and then processed. Hand separation requires the samples to be fresh and processed as soon as possible after harvest (Shaffer et al., 1990).

NIRS instruments use light at different wavelength in the NIR region to make measurements. The light is either absorbed or reflected by the sample and the wavelengths are determined at intervals between 700-2500 nm by a sensor. The absorption of the light in the NIRS region is primarily due to the frequency and arrangement of X-H bounds (Shenk and Westerhaus, 1994). Based on the absorption of light, the forage composition may be predicted. Calibration equations are developed from samples with known entities of interest (i.e acid detergent fiber or crude protein). These entities are associated with their spectral values. The most accurate calibration equations are developed from samples that represent all the variables that affect the NIRS. Shenk et al. (1979) demonstrated that the percent legume in grass-legume mixtures could be predicted within ± 10 % by NIRS.

Several researchers have attempted to use NIRS as an approach to determine botanical compositions. Some researchers have already proven the capability of NIRS to determine legume content in legume-grass mixtures (Petersen et al., 1987; Pitman et al., 1991). Coleman et al. (1985) concluded that with proper calibration (R^2 from 0.95 to 0.99 and SEP from 1.9 to 6.9), NIRS could accurately determine species composition. Petersen et al. (1987) reported a high R^2 when predicting species composition in tall fescue (*Festuca arundinacea* Schreb.) and white clover (*Trifolium repens* L) mixtures. Wachendorf et.al.(1999) successfully predicted red clover (*Trifolium pratense* L.) and white clover-grass mixtures content by using calibration equations from samples that included grass-clover mixtures collected at different years and locations. Coleman et al.

(1990) predicted the botanical composition of legumes and grasses mixtures using a calibration equation from a group of pure samples, each sample consisting of one of three botanical components to be validated by hand separation. They reported excellent prediction of the legume component with prediction coefficient of determination R^2 of 0.96 compared with caucasian bluestem *[Andropogon caucasicus* (Trin) C.E. Hubb] and cheatgrass (*Bromus tectorum* L.) that had R^2 of 0.77 and 0.84, respectively. Shaffer et al. (1990) predicted alfalfa (*Medicago sativa* L.) and ryegrass (*Lolium perenne* L.) in mixtures using an equation developed based on data from different locations and years. However, higher SEP was observed in prediction botanical composition of alfalfa-ryegrass from locations not included in the calibration equation. This may infer that calibration equations should be derived from samples representing all the variables that might influence the NIRS results, such as climate, soil type and species. Shaffer et al. (1990) concluded that approximately 200 samples were required to provide sufficiently accurate botanical composition prediction. Locher et al. (2005) obtained an accurate prediction of legume content over a broad variation of multispecies clover-grass, plant age, and site conditions.

It is generally believed that NIRS will accurately quantify the botanical composition of binary mixtures of vegetative grasses and legumes. However, no work has been done to develop calibration equations from several vegetative pasture grass-clover mixtures established at various latitudes over multiple- years period under North-central US growing conditions.

The objectives of this study were (i) to determine if NIRS could accurately predict species composition of several vegetative pasture grass-clover mixtures in three different locations over three-year period and (ii) to compare prediction accuracy of two calibration equations, one developed from pure laboratory-mixed samples and the other developed from hand separation samples, both clipped from a single location and year.

MATERIALS AND METHODS

Plant Material and Locations: Cool-season grass and clover binary mixtures were established in summer of 2001 at three Michigan State University (MSU) experimental stations: (i) Kellogg Biological Research Station at Hickory Corners (KBS) (42° 24' N, 85° 24' W); (ii) The Beef Cattle and Forage Research Station at Lake City (LC) (44°19' N, 85°12' W), and (iii) Upper Peninsula Experiment Station at Chatham (UPES) (46°33' N, 86°55' W). Three to five cutting events, depending on the location, were used to obtain samples during 2003-2005 growing seasons with 30 to 35 days interval between cutting events. More cuttings were taken at the KBS site since it is located further south than LC and UPES sites and has a longer growing season.

Twenty-four clover and grass cultivars were used for this study. Clovers included three red clover cultivars (VNS*, Star Fire, and Start), four white clover (KopuII, Ladino, Jumbo, and Alice), one kura clover cultivar (Endura). The grasses included two festuloliums cultivars (Duo and Hykor), four orchardgrass (Tekapo, Amba, Niva, and Sparta), seven perennial ryegrass (Aries, Maverick Gold, Quartet, Tonga, Barfort, Mara and Calibra) and three tall fescue cultivars (Bronson, K5666V, and Barolex).

At each location, the experiment was arranged in a randomized complete block design (RCBD) with three replications. Each replication consisted of 67 entries of different combinations of the above mentioned grass and clover cultivars seeded in 1.8 by 5 m plots.

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* Variety Not Stated

In addition, the grass and clover cultivars were established as monocultures in plots of the same size as these of the binary mixtures portion as previously explained in chapter 1.

Samples were hand clipped within a 0.25 m^2 quadrate in each plot when plants were 20 cm in height. Clipped samples were dried at 65 °C in a forced-air oven for 48 hr. All samples were ground to pass through a 1mm-screen in a Christy-Norris cyclone mill (Christy-Norris, Inc., Ipswitch, UK).

NIRS procedure: Two grams of each dried sample was packed into a sample holder with a 30 mm diameter quartz window and scanned with a NIRS monochromator (FOSS 6500). The wavelength range of 400-2500 nm was scanned at every 2 nm giving a total of 1050 data points. All the spectral data were recorded as log R^{-1}, where R is reflectance. Reflectance data were regresses on each constitute of the binary mixtures, that is, measured percentages of grass, legume and weed components to develop calibration equations. Standard deviation of calibration (SD), standard error of calibration (SEC) and calibration coefficient of determination (R^2) were used to evaluate the calibration equation.

The calibration equation was validated by comparing the reference standards (hand separated or artificially pure samples) with NIRS predicted values. Prediction coefficient of determination (R^2), standard error of prediction (SEP), slope, and bias were used as validation criteria. Calibration equations were developed using two different strategies described below.

I. Calibration Equation Developed From Artificially Pure Mixed Samples of 2004 Lake City data.

Pure grass and clover samples were clipped from monoculture plots at the Lake City site during the 2004 season. The samples were dried and ground as previously indicated. Artificially pure samples were hand mixed at 10% increments to a total of 4 g providing 11 possible combinations between each individual grass (Perennial ryegrass, orchardgrass, festuloliums and tall fescue) and clover species (white, red and kura clover). For instance, one mixture might contain 90% of perennial rye grass with 10% white clover, where a second mixture may include 80% of perennial rye grass with 20% white clover and so on). A total of 132 artificially mixed samples were made from the 4 grass and 3 clover species combinations. Mixed samples were packed into sample holders and scanned by NIRS as described previously. The calibration equations were developed (wavelengths 400 to 2492 nm) using an equation development program within the NIRS software from WinISI ver. 1.5 (FOSS NIRSystems, Inc., Eden Prairie, MN). The partial least-squares method was used to eliminate outliers. Five outliers were eliminated and remaining 127 samples were used to generate the calibration equation. Weeds were not included in the calibration equation.

II. Calibration Equation Developed From Hand-Separated Mixed Samples

In each location, a total of 201 (67 entries at three replications) samples were clipped from binary mixtures treatments and hand separated into three components: grasses, clovers, and undefined weeds species, mostly dandelion (*Taraxacum officinale).* The three separated components were dried, weighed separately, recombined and then ground using the same grinding method as the pure mixed samples. Each sample was scanned using NIRS following the previously described protocol. Two calibration equations were developed from hand-separated samples.

a. Calibration equation developed from hand-separation of 2004 Lake City data

This equation was derived from selected hand-separated samples from the 201 binary mixtures samples collected from LC during 2004 (weeds not included). Two outliers were eliminated from grass data and remaining 199 grass samples were used to develop the calibration equation. In addition, 60 outliers were eliminated from clover data and remaining 141 samples were used to generate the calibrating equation. The greater number of outliers eliminated from clover samples was due to the poor persistence of the clover over a period of years causing reduction in clover portion in the mixed samples (Chapter1). The purpose was to determine whether this calibration equation, which created from hand separation at single location and year, could provide a higher prediction accuracy of the binary mixtures composition than the calibration equation developed from artificial mixed samples obtained from the same single location and year. Validation statistics (R^2 and SEP) values obtained from artificial pure mixed and hand-separation equations were reported.

b. Calibration equation developed from multiple locations and years

This calibration equation was obtained based on the hand-separated mixtures samples collected from the three locations (KBS, LC and UPES) over 2003-2005 seasons. The reason for creating this equation was to determine if the prediction of the binary mixtures improved when more samples from different locations and years were included. A total of 1809 samples (201*3 locations *3 years) were clipped from the second cutting events in the three locations over three years. The samples were hand-separated as explained earlier. Half of the samples was randomly selected (904 samples) to create an equation to predict grass-clover species composition. The reflectance data from 471 grass, 455 clover and 381 weed samples were selected after outliers were eliminated using the Select program from WinISI, v. 1.5 software (Infrasoft International, LLC, Silver Springs, MD). Reflectance data were regressed on the proportion of each botanical component to develop three equations: one for grasses, one for clovers and one for weeds. All the equations were developed to predict the grass-clover botanical composition samples collected from all cutting events during 2003-2005 in three locations (KBS, LC, and UPES).

RESULTS AND DISCUSSION

I. Artificially pure mixed samples method: Table 1 lists the results of the NIRS calibration and validation statistics for estimating grass and clover content using the artificial pure mixed method. Data presented in Table 1 shows a very strong calibration equation with R^2 of 0.99 and standard error of calibration (SEC) of 1.4. The high R^2 of this equation is likely due to accurate handling of the pure mixtures samples with high precision in sample preparation and weighing samples using a sensitive scale. However, regression analyses of the spectral data with botanical composition resulted in prediction coefficient of determination (R^2) of 0.31 and 0.24 and SEP of 28.1 and 26.7 for grass and clover respectively (Table1). This indicated that this equation failed to predict the actual botanical composition of the pasture species. The reason for poor performance is that all artificially mixed samples were collected from one location and a single year. Additionally, the variation in plant maturity and weed content observed in the field samples were not represented in the lab mixed samples. Thus, the calibration equation developed from the artificially mixed samples of several grass and clover species collected from one location in a single year did not accuracy predict the species composition in field samples clipped from three different locations over three growing seasons. A higher prediction may obtain if the calibration equation was derived from samples representing all locations and years (Shaffer et. al., 1990).

II. Hand separation methods:

a. Equation developed from Hand-separation of LC 2004: Table 2 represents the calibration equation and validation statistics for predicting the botanical composition from an equation developed from hand separated samples collected in the single location and year. Calibration equations had R^2 of 0.97 for grass and 0.95 for clover. However, when this equation was applied to predict the botanical composition of samples collected from three locations and three years, similar prediction results were obtained to the results of the artificial pure mixed samples with validation R^2 of 0.25 for grass with somewhat improvement prediction for clovers with R^2 of 0.37 (Table 2). This clearly reflects the problem of obtaining enough representative samples that can include all spectra data from other locations.

b. Equation developed from multiple locations and years: Table 3 lists the calibration equations developed from hand-separated samples collected from three locations over three years. Calibration equations had R^2 of 0.82 for grass and 0.84 for both clovers and weeds (Table 3). The validation statistics for these equations resulted in an R^2 of 0.67 to 0.72, SEP of 6.9 to 12.8, and a bias of 0.3 to 0.6% (Table 3). The regression of each component is illustrated in Fig.1. Slopes were close to one for the grass and clover. Calibration samples comprised a wide range of values from 0 to 100 (g g^{-1} DM) for grass, 0 to 95 (g g^{-1} DM) for clover, and 0 to 85 (g g^{-1} DM) for weed (Fig1). Unlike the previous methods, this result suggests that using the same grass and clover species for both calibration and validation should improve the accuracy of NIRS predictions of these species. In spite of there being multiple species in the weeds portion, SEP for weeds was lower than grass and clover components. It is possible the weed species were more similar in reflectance values than the different species of grasses.

In conclusion, even though we had some success in predicting grass-clover mixtures based on multiple locations and years as compared with the equation created from a single location and year, this study did not result in as high R^2 as those observed in other studies (Locher et al., 2005). The lower R^2 observed in this study might be related to various factors. The calibration equations were derived from a large number of samples including different grass and clover cultivars in three different locations. Different cultivars, environmental conditions, the grinding processing and storing the samples can affect NIRS prediction (Shaffer, et al., 1990). In addition, clover and grass samples used in developing calibration equations were less mature than samples used in other studies. This may increase the difficulty of differentiation by NIRS since fiber content (hydrogen bonds arrangement and quantity) of immature grass and clover species were very similar. Coleman et al. (1985) indicated that the errors found with hay species were associated with (i) composition error (ii) machine error (iii) sampling error and (iv) methodology error. Hence, it is possible to generate a calibration equation developed from hand separation samples if the handing of the samples were controlled to reduce the variability among the samples.

CONCLUSIONS

Previously conducted research has shown success in predicting the botanical composition of grass-legume mixtures using artificial mixed pasture samples. This study showed that NIRS did not accuracy determine botanical composition of pasture grass-clover mixtures based on equation generated from artificial pure mixtures or hand-separated samples collected from a single location and year, since it may not represented the maturities of samples to be predicted. However, equations generated from hand separations of several grasses and clovers across all locations and years resulted in a more accurate prediction with some limitations. Some pasture grasses and clovers that are harvested at earlier maturities have similar fiber content (Chapter 2). This may explain the lower R^2 obtained in this study compared to other studies conducted using more mature hay samples. Environmental conditions may also cause variation in spectral properties of samples. Therefore, it is necessary to select diverse samples for calibration in order to obtain the best prediction from the pure sample equations.

This study, which presented two different strategies for developing a calibration equation, has shown that creating an equation from representative samples of a larger database of previously hand-separated mixtures can increase the prediction accuracy. Thus, NIRS can replace the hand separation method for botanical composition determination to a prediction accuracy (SEP) of ± 12%, and R^2 ranging from 0.67 to 0.72.

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Table1. Calibration and validation statistics for NIRS determination of botanical composition of grass-clover mixture samples collected over 2003-2005 in KBS, LC and UPES using calibration equations developed from artificial mixed samples of 2004 Lake City data.

Calibration					Validation				
Mixtures type	n+	SD‡	SEC‡‡	R^2¶	n	Bias	Slope	SEP§	R^2#
Grass	127	31.93	1.14	0.99	1440	-14.1	0.44	28.1	0.31
Clover	127	31.93	1.14	0.99	1375	6.7	0.40	26.7	0.24

Table2. Calibration and validation statistics for NIRS determination of botanical composition of grass-clover mixture samples collected over 2003-2005 in KBS, LC and UPES using calibration equations developed from hand-separation samples of 2004 Lake City data.

Calibration					Validation				
Mixtures type	n+	SD‡	SEC‡‡	R^2¶	n	Bias	Slope	SEP§	R^2#
Grass	199	19.13	3.22	0.97	1437	-20.81	0.36	27.41	0.25
Clover	141	20.53	4.17	0.95	1375	-0.88	0.56	20.95	0.37

Table.3. Calibration and validation statistics for NIRS determination of botanical composition of grass-clover mixture and weed samples collected over 2003-2005 in KBS, LC and UPES using calibration equations developed from the three years and three locations hand separation samples.

Calibration					Validation				
Mixtures type	n+	SD‡	SEC‡‡	R^2¶	n	Bias	Slope	SEP§	R^2#
Grass	471	21.8	9.3	0.82	946	0.53	0.90	12.8	0.67
Clover	455	22.1	8.9	0.84	902	0.35	0.94	12.22	0.72
Weed	381	10.0	4.0	0.84	763	0.64	1.04	6.99	0.68

+ Number of samples.
‡ Standard deviation of calibration sample set.
‡‡ Standard error of calibration sample set.
¶ Calibration coefficient of determination.
§ Standard error of prediction.
# Prediction coefficient of determination.

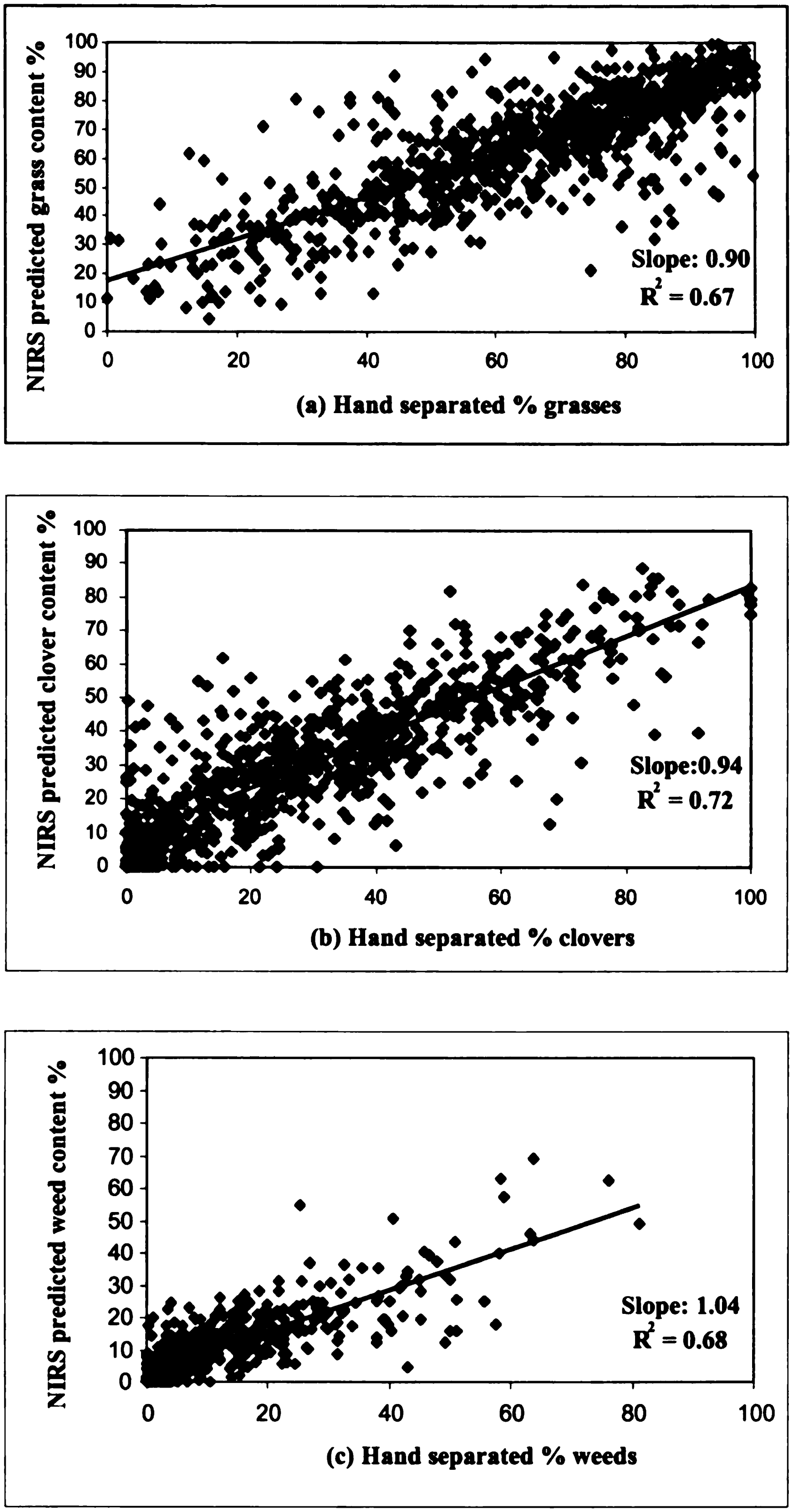

Fig1. Relationship of NIRS predicted percentage to hand-separated of grasses (a), clovers (b), and weeds (c) using a calibration equation of the hand separation of each component over three years at three experimental research stations. The solid line represents the regression with a slope and the coefficient of determination R^2

GENERAL SUMMARY

This research was conducted to evaluate the effects of growing pastures as binary mixtures of perennial grass and clovers upon animal preference, forage quality, plant persistence, botanical composition, and dry matter production.

Growing clovers with perennial grasses in binary mixtures resulted in increased pasture dry matter yield, persistence, forage quality, and animal preference. Grass monoculture treatments resulted in lower persistence and significantly more winter injury compared to the same grass species and cultivars grown in binary mixtures. Differences in persistence and winter injury due to location were observed. At northern latitudes (44°-46° N), where cold temperature predominate, higher incidence of winter injury occurred with less winter hardy cultivars. Tall fescue and orchardgrass cultivars resulted in better persistence than perennial ryegrass and festulolium. Orchardgrasses dry matter yield was slightly less than tall fescue but higher in animal preference. Among clover species, kura clover showed a high persistence at KBS and increased in clover content over time in all binary mixtures, which would indicate its potential as an excellent clover for grazing. Limited precipitation and high temperatures in 2005 can explain the reduction in pasture productivity at all locations.

There were significant differences in animal preferences among monoculture perennial grass species and cultivars. Forage quality of binary mixtures was higher in CP, and lower in ADF, and NDF than grass monoculture, which resulted in higher animal preferences for binary mixtures compared to monocultures of grass.

Perennial ryegrass-clover binary mixtures resulted in significantly higher forage quality and animal preference compared to perennial ryegrass monocultures. However, some perennial ryegrass cultivars that demonstrated a higher animal preference was less winter hardy. Binary mixtures of festulolium (Duo)-clover and perennial ryegrass-clover increased animal preference and forage quality compared to monoculture festulolium and perennial ryegrass, indicating these mixtures are a good choice for livestock producers to use for grazing. Total dry matter yield and animal preferences were not always positively correlated.

The study of Near-Infrared Reflectance Spectroscopy (NIRS) showed that using either pure mixed samples or hand separation from single location and year resulted in low prediction accuracy in determining grass-clover binary mixtures samples collected from KBS, LC and UPES locations during 2003-2005 growing season. In contrast, NIRS prediction accuracy was improved when the calibration equation was derived from hand-separated samples collected from the same locations and years.

In summary, this research provided a better understanding of the importance of species and cultivar selection for performance in grazing systems across three latitudes in Michigan. It also provided helpful information on grass-clover relationships in pastures over time under rotational grazing. In addition, the study documented the positive benefits of proper grass-clover cultivar and species selection for higher animal preference and plant persistence. Finally, NIRS has the potential to replace hand-separation in determining pasture species composition when calibration equation was developed from a large data set, which represents environments between locations and years.

APPENDICES

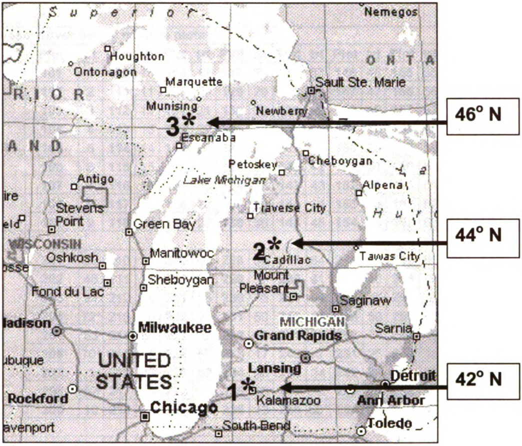

Fig 1A. Michigan map showing the three experimental locations representing different latitudes in Michigan: Kellogg Biological Research Station at Hickory Corners **(KBS)** (1), Lake City Research Station **(LC)** (2), and Upper Peninsula Experiment Station at Chatham **(UPES)** (3).

Grass-clover binary mixture plots																
	Plot	**trt**	**Plot**	**trt**	**Plot**	**trt**	**Plot**	**trt**	**Plot**	**trt**	**Plot**	**trt**	**Plot**	**trt**	**Plot**	**trt**
	101	1	**110**	10	**119**	19	**128**	28	**137**	37	**146**	46	**155**	55	**164**	64
	102	2	**111**	11	**120**	20	**129**	29	**138**	38	**147**	47	**156**	56	**165**	65
	103	3	**112**	12	**121**	21	**130**	30	**139**	39	**148**	48	**157**	57	**166**	66
	104	4	**113**	13	**122**	22	**131**	31	**140**	40	**149**	49	**158**	58	**167**	67
R1	**105**	5	**114**	14	**123**	23	**132**	32	**141**	41	**150**	50	**159**	59	**168**	68*
	106	6	**115**	15	**124**	24	**133**	33	**142**	42	**151**	51	**160**	60	**169**	69*
	107	7	**116**	16	**125**	25	**134**	34	**143**	43	**152**	52	**161**	61	**170**	70*
	108	8	**117**	17	**126**	26	**135**	35	**144**	44	**153**	53	**162**	62	**171**	71*
	109	9	**118**	18	**127**	27	**136**	36	**145**	45	**154**	54	**163**	63	**172**	72*
	201	56	210	31	**219**	63	**228**	36	**237**	64	**246**	43	**255**	61	**264**	55
	202	48	211	27	**220**	29	**229**	9	**238**	41	**247**	40	**256**	3	**265**	35
	203	21	212	39	**221**	18	**230**	15	**239**	54	**248**	37	**257**	2	**266**	57
R2	**204**	72*	213	16	**222**	26	**231**	50	**240**	11	**249**	59	**258**	47	**267**	4
	205	53	214	60	**223**	8	**232**	30	**241**	22	**250**	14	**259**	49	**268**	65
	206	70*	215	44	**224**	52	**233**	17	**242**	6	**251**	25	**260**	45	**269**	38
	207	1	216	33	**225**	62	**234**	23	**243**	34	**252**	24	**261**	51	**270**	69*
	208	67	217	58	**226**	5	**235**	66	**244**	12	**253**	20	**262**	42	**271**	71*
	209	10	218	28	**227**	7	**236**	13	**245**	68*	**254**	32	**263**	46	**272**	19
	301	7	**310**	17	**319**	50	**328**	45	**337**	35	**346**	23	**355**	3	**364**	67
	302	28	**311**	27	**320**	12	**329**	71*	**338**	43	**347**	47	**356**	46	**365**	44
	303	58	**312**	59	**321**	53	**330**	72*	**339**	18	**348**	24	**357**	48	**366**	22
R3	**304**	70*	**313**	15	**322**	6	**331**	20	**340**	40	**349**	9	**358**	51	**367**	32
	305	2	**314**	42	**323**	63	**332**	19	**341**	38	**350**	57	**359**	8	**368**	68*
	306	54	**315**	69*	**324**	39	**333**	62	**342**	60	**351**	25	**360**	16	**369**	10
	307	31	**316**	4	**325**	1	**334**	29	**343**	30	**352**	11	**361**	61	**370**	37
	308	52	**317**	21	**326**	33	**335**	34	**344**	49	**353**	65	**362**	13	**371**	56
	309	14	**318**	66	**327**	41	**336**	5	**345**	26	**354**	55	**363**	64	**372**	36

Clover plots

R1	**101**	73	**102**	74	**103**	75	**104**	76	**105**	77	**106**	78	**107**	79	**108**	80
R2	**201**	79	**202**	77	**203**	75	**204**	80	**205**	78	**206**	73	**207**	74	**208**	76
R3	**301**	76	**302**	78	**303**	74	**304**	73	**305**	80	**306**	79	**307**	77	**308**	75

Grass plots

R1	**101**	81	**103**	83	**105**	85	**107**	87	**109**	89	**111**	91	**113**	93	**115**	95
	102	82	**104**	84	**106**	86	**108**	88	**110**	90	**112**	92	**114**	94	**116**	96
R2	**201**	89	**203**	83	**205**	85	**207**	93	**209**	88	**211**	81	**213**	96	**215**	95
	202	87	**204**	94	**206**	90	**208**	92	**210**	86	**212**	82	**214**	91	**216**	84
R3	**301**	96	**303**	92	**305**	89	**307**	87	**309**	82	**311**	90	**313**	85	**315**	84
	302	91	**304**	88	**306**	95	**308**	94	**310**	93	**312**	86	**314**	81	**316**	83

Fig. 2 A. The design map of grass-clover binary mixtures and grass and clover monoculture treatments. (* Trt.# 68,69,70,71 and 72 in the binary mixtures plots are blanks)

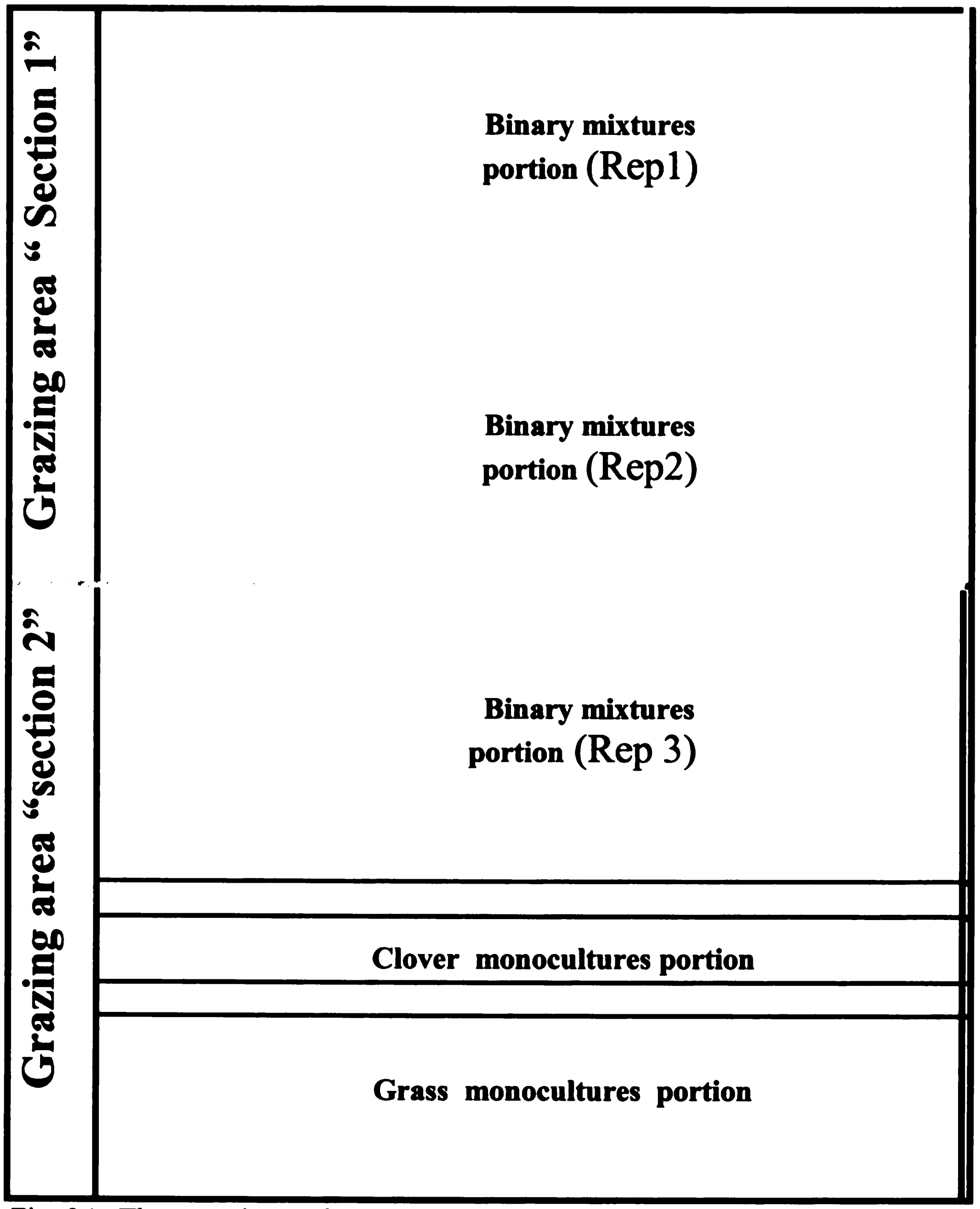

Fig. 3A. The experimental area grazing arrangement.

Table1 A. Soil type, soil pH, cutting schedule over 2004 and 2005 growing seasons at Kellogg Biological Research Station (KBS), Lake City (LC) and Upper Peninsula Experiment Station (UPES) and the grass and clover cultivars established in this study.

Loc	Soil Type	pH	Cutting Schedule 2004	Cutting Schedule 2005	Grasses Species	Grasses Cultivars	Clover Species	Clover Cultivars
KBS	Kalamazoo loam.	6.6	24 –May	4- May	Festuloliums	Duo	Kura clover	Endura
	Fine-loamy, mixed,		24 –June	14-June	Festuloliums	Hykor		
	semiactive, mesic		26-July	2-Aug.				
	Typic Hapludalfs		9-Sept	13-Oct.	Orchardgrasses	Tekapo	Red clover	Star Fire
LC	Nester sandy loam	6.1	19-May	23- May	Orchardgrasses	Amba	Red clover	Start
	(Fine, mixed,		21- June	7- July	Orchardgrasses	Niva	Red clover	VNS*
	semiactive, frigid		5- Aug	9- Aug	Orchardgrasses	Sparta		
	Oxyauic Glossudalfs)		7- Sept	2-Oct				
UPES	Stony loam (Coarse-	7.6	7-June	1- June	Perennial ryegrass	Aries	White clover	Kopu II
	loamy, mixed, frigid		12-July	10- July	Perennial ryegrass	Mavrck Gold	White clover	Alice
	Typic Haplorthods		22-Aug.	23- Aug	Perennial ryegrass	Quartet	White clover	Jumbo
			17-Oct.	29-Sept	Perennial ryegrass	Tonga	White clover	ladino
					Perennial ryegrass	Barfort		
					Perennial ryegrass	Mara		
					Perennial ryegrass	Calibra		
					Tall fescues	Bronson		
					Tall fescues	K5666V		
					Tall fescues	Barolex		

* Variety Not Stated

Table 2 A. Total monthly precipitation (mm) at Kellogg Biological Research Station (KBS), Lake City (LC) and Upper Peninsula Experiment Station (UPES) during the growing season of 2004-2005 comparing with 30-year average.

KBS			
Month of growing season	2004	2005	30-yr Ave.
	------------------- mm ---------------------		
April	19.8	6.3	97.0
May	248.4	50.8	89.0
June	85.2	124	98.0
July	76.1	122.4	93.7
Aug.	131	11.4	100.1
Sep.	44.1	69.9	108
Total	**604.6**	**384.8**	**585.8**
Seasonal Average	100.8	64.2	97.7
LC			
Month of growing season	2004	2005	30-yr Ave.
	------------------- mm ---------------------		
April	100.6	26.4	70.4
May	161.3	65.5	70.8
June	61.7	52.1	75.0
July	46.7	93.7	72.6
Aug.	53.0	113.8	92.9
Sep.	21.8	103.1	94.5
Total	**445.1**	**454.6**	**476.2**
Seasonal Average	74.2	75.8	79.4
UPES			
Months of growing season	2004	2005	30-yr Ave.
	------------------- mm ---------------------		
April	60.7	36.8	40.13
May	107.4	51.0	72.4
June	75.9	41.4	77.7
July	92.9	98.5	90.6
Aug.	145.8	61.4	78.1
Sep.	50.5	123.4	93.7
Total	**533.2**	**412.5**	**452.6**
Seasonal Average	88.9	68.8	75.5

Table 3 A. Average daily maximum air temperatures (°C) at Kellogg Biological Research Station (KBS), Lake City (LC) and Upper Peninsula Experiment Station (UPES) during the growing season of 2004-2005 comparing to 30-year average.

	KBS		
Months of growing season	**2004**	**2005**	**30-yr Ave.**
	----------------° C -----------------		
April	17.3	18.2	15.3
May	22.0	19.9	22.3
June	25.2	30.0	27.1
July	27.5	30.4	29.2
Aug.	25.7	29.4	27.9
Sep.	26.7	27.3	23.8
	LC		
Months of growing season	**2004**	**2005**	**30-yr Ave.**
	----------------° C -----------------		
April	13.1	14.8	11.4
May	17	16.4	19.3
June	22.7	27.5	24.2
July	25.5	27.4	26.6
Aug.	23.5	26.0	25.1
Sep.	24.9	23.8	20.3
	UPES		
Months of growing season	**2004**	**2005**	**30-yr Ave.**
	----------------° C -----------------		
April	7.7	13.0	10.4
May	15.4	15.7	18.9
June	19.9	25.5	24.3
July	23.6	27.2	25.6
Aug.	22.4	26.4	24.9
Sep.	23.3	23.3	20.6

Table 4 A. Average daily minimum air temperatures (°C) and snow fall (cm) at Kellogg Biological Research Station (KBS), Lake City (LC) and Upper Peninsula Experiment Station (UPES) from January through April of 2003-2006 comparing with 30-yr. average

KBS										
	2003		2004		2005		2006		30-yr Ave.	
Month	Temp °C	Snow cm	Temp °C	Snow cm	Temp °C	Snow cm	Temp °C	Snow cm	Temp °C	Snow cm
Jan	-9.8	18.9	-8.4	30.09	-9	17.6	-2	19.8	-8.8	20.5
Feb	-10.1	10.4	-7.7	11.6	-5	6.4	-6	9.2	-8.1	13.5
Mar	-3.2	1.7	0.2	1.5	-5	1.2	-2	4.2	-3.2	10.2
Apr	3.3	0.5	2.3	0	4	0	4	0.3	2.4	2.9
L C										
Jan	-12.6	18	-14	19	-13	15	-5	14	-13	19.3
Feb	-15.4	13	-11	12	-8	14	-11	16	-12.8	15.4
Mar	-9.3	12	-4	11	-10	11	-6	13	-7.9	12.2
Apr	-2.3	4	0	2	0	5	0	2	-0.9	4.4
UPES										
Jan	-15.6	35	-15.8	32	-14	37	-7	31	-12.7	35.9
Feb	-18.4	26	-14.0	29	-10	24	-13	28	-12.4	29
Mar	-12.8	28	-7.4	22	-10	21	-6	22	-8.3	18.9
Apr	-5.8	10	-2.7	8	-1	11	0	7	-2.3	6.6

Table.5 A. Average animal weight (kg), and the number of animals in each grazing event during 2004-2006 grazing seasons.

	2004				**2005**			**2006***
Animal Info	28-May	26-Jun	28-Jul	9-Sep	6-May	16-Jun	4-Aug	12-May
Ave animal weight (kg)	**266.22**	**311.85**	**384.42**	**411.11**	**243.46**	**365.60**	**403.10**	**443.72**
Number of Animals used	**10**	**4**	**4**	**3**	**6**	**3**	**3**	**16**
Breed: Holstein	Steers	Steers	Steers	Steers	Steers	Steers	Steers	Heifers

* Single grazing was taken during 2006 season

Table 6 A. ANOVA for winter injury, spring and, fall ground cover and spring and fall clover for binary mixture data from years 2004-2006, locations (KBS, LC and UPES) and 67 binary mixtures(treatments).

		WI[¶]		SGC[*]		FGC[x]		SC[y]		FC[z]	
Effect	DF	F-value	Pr > F	F-value	Pr > F	F-value	Pr > F	F-value	Pr > F	F-value	Pr > F
year	2	23.49	<.0001	15.61	0.0002	1.69	0.2619	71.18	<.0001	9.57	0.0299
loc	2	52.98	0.0002	90.93	<.0001	3.86	0.0837	36.39	0.0004	9.57	0.0299
year*loc	4	35.63	<.0001	32.85	<.0001	8.52	0.0177	64.99	<.0001	232.46	0.0001
trt	66	21.88	<.0001	32.08	<.0001	11.69	<.0001	11.35	<.0001	7.74	0.0001
year*trt	132	3.16	<.0001	3.87	<.0001	4.00	<.0001	5.29	<.0001	4.36	0.0001
loc*trt	132	6.82	<.0001	6.14	<.0001	1.62	0.0002	5.21	<.0001	3.38	0.0001
year*loc*trt	264	2.74	<.0001	3.37	<.0001	1.71	<.0001	2.56	<.0001	2.54	0.0001

¶ WI= Winter Injury
*SGC= Spring Ground Cover
xFGC= Fall Ground Cover (2006 data is not included)
ySC= Spring Clover
zFC= Fall Clover (2006 data is not included)

Table 7 A. ANOVA for winter injury, spring, and fall ground cover for grass monoculture data from years 2004-2006, locations (KBS, LC and UPES) and 16 grass varieties (Cultivars)

		WI¶		SGC*		FGC^x	
Effect	DF	F-value	Pr > F	F-value	Pr > F	F-value	Pr > F
year	2	121.83	<.0001	83.55	<.0001	9.19	0.0387
loc	2	152.05	<.0001	171.50	<.0001	13.93	0.0158
year*loc	4	133.52	<.0001	125.36	<.0001	49.22	0.0022
var	15	50.09	<.0001	43.27	<.0001	30.55	<.0001
year*var	30	2.23	<.0007	1.98	<.0034	2.85	<.0022
loc*var	30	8.74	<.0001	5.30	<.0001	4.97	<.0001
year*loc*var	60	4.47	<.0001	4.42	<.0001	4.02	<.0001

¶ WI= Winter Injury

*SGC= Spring Ground Cover

xFGC= Fall Ground Cover (2006 data is not included)

Table 8 A. ANOVA for winter injury, spring, and fall ground cover for clover monoculture data from years 2004-2006, locations (KBS, LC and UPES) and eight clover varieties (Cultivars).

		WI[x]		SGC[y]		FGC[z]	
Effect	DF	F-value	Pr > F	F-value	Pr>F	F-value	Pr>F
year	2	78.68	<.0001	9.57	0.0059	0.01	0.9381
loc	2	70.25	<.0001	26.32	0.0011	32.13	0.0006
year*loc	4	27.33	<.0001	21.99	0.0002	2.86	0.1343
var	7	6.67	0.0001	25.77	<.0001	26.32	<.0001
year*var	14	5.55	0.0004	12.53	<.0001	2.17	0.0682
loc*var	14	2.73	0.0114	5.84	<.0001	3.79	0.0013
year*loc*var	28	3.74	0.0055	5.85	0.0003	4.17	0.0029

x WI= Winter injury.
y SGC =Spring ground cover
zFGC =Fall ground cover (2006 data is not included)

Table 9 A. ANOVA for dry matter yield, animal preference and forage quality (CP, ADF and NDF) for binary mixture data from years 2004-2005, locations (KBS, LC and UPES), cutting and 67 binary mixtures(treatments).

		Yield		*Animal pre		CP[x]		ADF[y]		NDF[z]	
Effect	DF	F-value	Pr > F	F-value	Pr>F	F-value	Pr>F	F-value	Pr>F	F-value	Pr>F
Year	1	0.00	0.9971	133.02	<.0001	120.29	<.0001	0.28	0.6134	6.05	0.0491
loc	2	2.09	0.2049	NA	NA	152.96	<.0001	73.49	<.0001	20.88	0.0020
year*loc	2	19.49	0.0024	NS	NA	5.54	0.0433	15.57	0.0042	6.40	0.0326
trt	66	4.92	<.0001	3.89	<.0001	17.94	<.0001	23.02	<.0001	-	<.0001
year*trt	66	1.78	0.0005	1.86	<.0001	3.46	<.0001	3.79	<.0001	4.52	<.0001
loc*trt	132	2.01	<.0001	NA	NA	2.96	<.0001	1.99	<.0001	2.46	<.0001
year*loc*trt	132	1.96	<.0001	NA	NA	1.94	<.0001	1.99	<.0001	2.16	<.0001
cut	4	91.19	<.0001	91.50	<.0001	359.62	<.0001	7624.24	<.0001	-	<.0001
year*cut	4	61.35	<.0001	103.82	<.0001	546.25	<.0001	810.20	<.0001	206.92	<.0001
loc*cut	8	44.13	<.0001	NA	NA	246.22	<.0001	101.93	<.0001	52.83	<.0001
year*loc*cut	8	85.81	<.0001	NA	NA	15.85	<.0001	9.52	<.0001	3.99	0.0076
cut*trt	264	3.12	<.0001	1.94	<.0001	2.49	<.0001	2.27	<.0001	-	<.0001
year*cut*trt	264	2.39	<.0001	1.92	<.0001	1.56	<.0001	1.74	<.0001	1.77	<.0001
loc*cut*trt	528	1.69	<.0001	NA	NA	1.21	0.0060	1.54	<.0001	1.20	0.0080
year*loc*cut*trt	528	1.77	<.0001	NA	NA	1.40	0.0003	1.69	<.0001	1.56	<.0001

*Animal pre= Animal preferences (Animal preferences conducted at KBS only)
xCP = Crude protein
yADF = Acid detergent fiber.
z NDF = Neutral detergent fiber.
NA = Data is not applicable.

Table 10.A. ANOVA for dry matter yield, animal preference and forage quality (CP, ADF and NDF) for grass monoculture data from years 2004-2005, locations (KBS, LC and UPES), cutting and 16 grass varieties (Cultivars).

		Yield		*Animal pre		CP[x]		ADF[y]		NDF[z]	
Effect	DF	F-value	Pr > F	F-value	Pr>F	F-value	Pr>F	F-value	Pr>F	F-value	Pr>F
year	1	88.00	<.0001	94.36	<.0001	445.81	<.0001	0.28	0.6176	1436.09	<.0001
loc	2	10.94	0.0100	NA	NA	36.41	0.0004	39.34	0.0004	63.37	<.0001
year*loc	2	3.96	0.0801	NA	NA	162.53	<.0001	15.01	0.0046	7.07	0.0264
var	15	12.34	<.0001	4.08	0.0005	8.10	<.0001	21.05	<.0001	18.93	<.0001
year*var	15	0.87	0.6001	3.95	<.0001	2.06	0.0196	8.60	<.0001	4.75	<.0001
loc*var	30	2.73	0.0001	NA	NA	1.70	0.0291	2.62	0.0003	1.40	0.1138
year*loc*var	30	1.78	0.0195	NA	NA	2.03	0.0056	2.13	0.0033	3.37	<.0001
cut (Graze)	4	31.85	<.0001	129.17	<.0001	39.19	<.0001	177.40	<.0001	460.23	<.0001
year*cut	4	29.44	<.0001	1.85	0.1762	163.51	<.0001	45.48	<.0001	169.04	<.0001
loc*cut	8	16.10	<.0001	NA	NA	83.50	<.0001	102.39	<.0001	125.07	<.0001
year*loc*cut	8	19.20	<.0001	NA	NA	8.95	<.0001	67.43	<.0001	46.12	<.0001
cut*var	60	2.55	<.0001	4.70	<.0001	2.49	<.0001	2.91	<.0001	2.25	<.0001
year*cut*var	60	1.62	0.0082	5.15	<.0001	1.91	0.0006	1.76	0.0024	2.38	<.0001
loc*cut*var	120	0.91	0.6951	NA	NA	1.74	0.0002	1.67	0.0004	1.63	0.0008
year*loc*cut*var	120	1.12	0.2737	NA	NA	1.70	0.0030	1.96	0.0002	1.37	0.0547

*Animal pre= Animal preferences (Animal preferences conducted at KBS only)
xCP = Crude protein
yADF = Acid detergent fiber.
z NDF = Neutral detergent fiber.
NA = Data is not applicable.

Table 11 A. ANOVA for dry matter yield, animal preference and forage quality (CP, ADF and NDF) for clover monoculture data from years 2004-2005, locations (KBS, LC and UPES), cutting and eight clover varieties (Cultivars).

		Yield		*Animal pre		CP[x]		ADF[y]		NDF[z]	
Effect	DF	F-value	Pr > F	F-value	Pr>F	F-value	Pr>F	F-value	Pr>F	F-value	Pr>F
year	1	317.87	<.0001	55.14	0.0018	1.55	0.2589	161.66	<.0001	13.60	0.0102
loc	2	30.66	0.0007	NA	NA	495.23	<.0001	224.99	<.0001	146.52	<.0001
year*loc	2	47.89	0.0002	NA	NA	14.31	0.0052	7.60	0.0227	4.33	0.0687
var	7	2.19	0.0548	0.57	0.7664	8.70	<.0001	19.16	<.0001	9.30	<.0001
year*var	7	4.39	0.0010	1.97	0.1328	3.84	0.0048	0.81	0.5896	0.77	0.6149
loc*var	14	2.31	0.0186	NA	NA	0.97	0.5070	0.86	0.6032	0.49	0.9190
year*loc*var	14	3.69	0.0005	NA	NA	1.63	0.1756	3.17	0.0168	2.32	0.0606
cut (Graze)	4	10.17	<.0001	11.32	<.0001	68.94	<.0001	208.74	<.0001	152.23	<.0001
year*cut	4	9.96	<.0001	3.35	0.0711	33.98	<.0001	24.50	<.0001	31.04	<.0001
loc*cut	8	8.77	<.0001	NA	NA	26.02	<.0001	12.36	<.0001	9.27	<.0001
year*loc*cut	8	12.67	<.0001	NA	NA	5.34	0.0015	4.82	0.0030	9.29	<.0001
cut*var	28	2.21	0.0007	1.41	0.1211	1.60	0.0367	2.79	<.0001	1.76	0.0159
year*cut*var	28	1.30	0.1765	0.53	0.8118	0.60	0.8490	0.78	0.6818	0.66	0.7967
loc*cut*var	65	1.17	0.2513	NA	NA	1.13	0.3101	0.99	0.4794	0.72	0.8415
year*loc*cut*var	65	1.66	0.0374	NA	NA	1.92	0.0930	4.37	0.0009	3.48	0.0051

*Animal pre= Animal preferences (Animal preferences conducted at KBS only)
xCP = Crude protein
yADF = Acid detergent fiber.
z NDF = Neutral detergent fiber.
NA = Data is not applicable.