YIELD AND NUTRITIONAL VALUE OF ORCHARDGRASS AS INFLUENCED BY NITROGEN AND SULPHUR FERTILIZATION AND ASSOCIATED RED CLOVER

by

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TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

INTRODUCTION

Orchardgrass (Dactylis glomerata, L.) is a well adapted cool season, perennial grass in the temperate areas of the United States and its value as a herbage plant with regard to its quality for the feeding of livestock is well known. Orchardgrass responds well to high nitrogen (N) fertilization, is competitive and tolerates intensive cutting or grazing management. However, studies on forages have shown that responses to heavy N fertilization were often limited by a deficiency of sulphur (S) (Stewart and Porter, 1967; Aulakh et al., 1976). An insufficient supply of S can influence both the yield and quality of the herbage produced. Under conditions of high N fertilization and S deficiency, protein synthesis is reduced by a lack of the S containing amino acids, cysteine and methionine (Dijkshoorn et al., 1960), and this is reflected by marked accumulation of N in the plant as nitrate-nitrogen, amides and free amino acids (Terman et al., 1976; Goh and Kee, 1978). Such herbages are nutritionally unbalanced and can cause nutritional disorders when fed to ruminants. Thus, the ratio of total N to total S becomes important as a measure of feeding value of the forage (Tisdale, 1977).

The effect of S on forage quality is not as well documented as its effects on dry matter yields. Several studies have shown a yield response to added S in the second year (Beaton, 1966; Brupbacher and Sedberry, 1965). Metson (1973) reported that S responses of grasses in grass-legume mixtures are associated with the rate of N application. This response of grasses in grass-legume mixtures has been attributed to the early removal of S by their legume components where legumes begin growth earlier in the season and exhaust the available supply, thus limiting the available S for the grasses (Metson, 1973). However, legumes can be used successfully to meet the N requirements of grasses as well as to improve the quality of grass-legume mixtures (Mulder, 1952; Cowling, 1961; Carter and School, 1962; Blaser et al., 1969; Sau, 1970; Hunt et al., 1975; Johanson and Kerridge, 1979). The beneficial effects of legumes as components in forage mixtures is due to the ability of symbiotic Rhyzobium bacteria to fix atmospheric N for growth and improved protein content.

Review of Literature

Nitrogen Fertilization of Forages

Effect on Dry Matter Yields. Beneficial effects of N application in increasing forage dry matter yields are well documented. Sauerbery (1975) reported that when an orchardgrass pasture was fertilized with 0, 80, 160, 320 or 480 kg N/ha either in a single or four split applications, dry matter yields increased in all years with increasing levels of N. Kunelius et al. (1977) stated that the application of N (99 to 495 $kg \cdot ha^{-1} \cdot yr^{-1}$) to orchardgrass resulted in significant increases in dry matter yields whereas, Mouchel et al. (1977) reported that the application of more than 360 kg N/ha is of little benefit to the grass. Chung (1978) observed that the optimum fertilizer rates for dry matter yields of orchardgrass and native grass were 200 to 300 kg N/ha.

Pure stands of grass respond readily to applied N. Cucu et al. (1973) reported that the dry matter yields of orchardgrass increased with increasing amounts of applied N from 300 to 660 kg N/ha whereas, Sillanpaa and Rinne (1975) and Reid (1970) showed that response to N fertilization peaked at 300 kg N/ha and declined thereafter for orchardgrass and perennial ryegrass (Lolium perenne, L.), respectively. Noller and Rhykerd (1974), working with orchardgrass, reported that the dry matter yields more than trebled as N rate was increased from 0 to 600 kg/ha and declined slightly thereafter. Similar conclusions were made by Donohue (1975), who found that herbage yields of orchardgrass increased with increasing rate of applied N up to 600 kg/ha, remained fairly constant between 600 and 1500 kg/ha and decreased markedly at 1800 and 2100 kg/ha. This decrease in dry matter yield with very high levels of N fertilization may be due to increased plant nitrate-nitrogen contents. Nitrate-nitrogen concentration reached toxic levels if the recommended rates of 400 to 450 kg N/ha in the first year and 500 to 550 kg N/ha in the second year were exceeded (Moga et al., 1973). Large applications of N may also result in excessive soil acidity, accompanied by sharp reductions in exchangeble bases, reduced phosphorous (P) availability, and sharp increases in soluble aluminum and manganese (Heath et al., 1978).

Nutrient Composition and Digestibility. It is well known that crude protein content of forages increases with increasing levels of fertilizer N (Lampeter et al., 1973; Cucu et al., 1977; Wilman and Wright, 1978). Kunelius and Suzuki (1977) obtained a linear increase in total N concentration as fertilization was increased from 99 to 495 kg N/ha/yr in orchardgrass, whereas, Noller and Rhykerd (1974) reported that the crude protein and nitrate-nitrogen contents plateaued at 600 kg N/ha. According to Donohue (1975), content of total N and nitrate-nitrogen in the herbage increased with increasing N up to 1200 kg/ha and remained the same or was lower at higher levels. Dotzenko (1961) obtained protein levels ranging from 10 to 18 percent in orchardgrass with N rates of 90 to 716 kg/ha.

Nitrate in forages above certain levels can be toxic to animals. Nitrate-nitrogen content of .2 to .3% has been reported as the critical range for forage where it is the only source of feed (Gillingham et al., 1969; Hallock et al., 1973). Moga et al. (1973) reported that nitrate-nitrogen contents of orchardgrass reached toxic levels only when the recommended N application rates of 400 to 450 kg/ha in the first year and 500 to 550 kg/ha in the second year were exceeded.

The effects of N fertilization on the concentration of other mineral elements have been inconsistent. While some have reported no effect of applied N on the concentration of magnesium (Mg) in forage (Gorlach et al., 1973; Firek, 1974; Korter, 1974), others have noted an increase (Reid et al., 1964; Stuzynska, 1974; Reid et al., 1974; Sillanpa et al., 1975; Hojjati et al., 1977).

According to several authors (Reid et al., 1966; Gorlach and Curylo, 1973; Reid et al., 1974), applied N had no effect on the concentration of calcium (Ca) in forage.

However, Koter (1974) reported a decrease in Ca content of orchardgrass as N fertilization was increased from 120 to 360 kg/ha. To the contrary, Stuzynska (1974) and Sillanpaa and Rinne (1975) obtained an increase in Ca content with increasing levels of fertilizer N in orchardgrass.

O'Connor and Vartha (1969) and Goh and Kee (1978) reported a significant decrease in S content in orchardgrass and perennial ryegrass with increasing N applications, whereas, Jones (1960), Lancaster et al. (1971) and Burmester et al. (1981) reported an increase in S content in orchardgrass and tall fescue with increasing levels of N application.

Variable results have been reported on the effects of N fertilization on in vitro and in vivo dry matter digestibility of protein and fiber components. Poulton et al. (1957) observed that crude protein digestibility of orchardgrass was increased as N fertilization was increased from 112 to 224 kg/ha, whereas, no effects were seen beyond 224 kg/ha. They also reported that fiber digestibility was not influenced by N fertilization. Bratzler et al. (1959) fertilized orchardgrass with N up to 336 kg/ha and found that the digestibility of fiber was not altered. However, protein digestibility was increased. Reid et al. (1966) reported that N had little effect on apparent digestibility of dry matter or cellulose in the first cut of orchardgrass hay when fed to sheep.

Wilman (1978) observed that application of N increased the proportion of crude protein more than it reduced the proportion of water soluble carbohydrates and it increased the proportion of cell content. He concluded that the increase in proportion of cell content was sufficient to explain the positive effect of N application on digestibility. Daughtry et al. (1978) reported that leaves of orchardgrass fertilized with 112 kg N/ha contained 51.1% cell wall constituents (CWC) and 29.5% hemicellulose as compared to 58.1% CWC and 24% hemicellulose in leaves of non-fertilized grass.

Nitrogen fertilization increased in vitro dry matter digestibility (Reid et al., 1966; Belasko, 1977; Goh and Kee, 1978). However, Lampetor et al. (1973) showed little or no effect on dry matter digestibility of orchardgrass, when fertilizer N was increased from 100 to 400 kg/ha.

Animal Parameters. Blaser (1964) reported that the application of N fertilizer increased the carrying capacity (as measured by weight gain or milk production per hectare) and protein content of forages, however, individual animal performance was usually not changed. Reid et al. (1973) also showed that levels of N up to 504 $kg \cdot ha^{-1} \cdot yr$ had no effect on the live weight of ewes maintained on orchardgrass. High levels of N fertilizer do not usually have a deleterious effect on animal performance (Blaser, 1964; Degroot, 1970). However, health problems may arise from the use of high levels of fertilizer N due to nitrate poisoning, an imbalance of N to S ratio, protein to energy ratio and other nutrients (Heath et al., 1978).

Seidler et al. (1972) showed when orchardgrass was fertilized with 320 or 640 kg N/ha, digestibility and N balance were higher for the orchardgrass hay fertilized with the high rate of N. Reid et al. (1974) maintained ewes on orchardgrass forage fertilized with N up to 504 kg/ha and found no significant effect on serum levels of Ca and Mg. Carver et al. (1978) observed an increase in plasma urea N levels of steers with increasing levels of N fertilization to forages.

Sulphur Fertilization of Forages

Effect On Dry Matter Yields. The application of S increased yield of forages in soils deficient in S (Bledsoe et al., 1947; Cairns and Carson, 1961; Goh and Kee, 1978) and yield responses under field conditions have been reported in at least 33 states in the United States and in parts of Canada (Tisdale, 1977). The responses of grasses to S appears to vary in magnitude depending on the species studied (O'Connor, 1961; Vartha, 1963; O'Connor and Vartha,

1969) and on the chemical form in which S is applied (Cairns and Carson, 1961). Sodium sulphate was more effective than gypsum, and gypsum was more effective than elemental S in the year of application (Cairns and Carson, 1961). O'Connor and Vartha (1969) also found that the response to gypsum was significantly greater than to comparable levels of elemental S.

Lancaster et al. (1971) reported that the dry matter yield of orchardgrass was significantly increased when 10, 20 or 40 ppm S was applied as sodium sulphate, but no significant response was noted for the legumes. However, others (Oke, 1969; Gilbert et al., 1981) have reported that S application significantly increased the legume yield. Jones (1962) reported that the first 22 kg of S/ha increased yield of subterraneum clover (Trifolium subterraneum L.), but rates beyond 22 kg S/ha had no effect.

Several authors have reported that the response to S by orchardgrass is dependent on the level of N fertilization (Jordan et al., 1958; Jordan et al., 1960). O'Connor and Vartha (1969) observed that the dry matter yield of orchardgrass was increased with increased S supply and the highest level of N fertilization. Application of S had no effect on the dry matter yield in the absence of added N. Sorenson et al. (1968) and Stewart and Porter (1969) also reported that the addition of S alone did not have any effect on the yield of alfalfa (Medicago sativa, L.) and wheat (Triticum aestivum, L.), respectively. Scattini (1981) reported that native pastures responded to fertilizer containing S by increasing the yields at high N rates. Burmester et al. (1981), found that S fertilizer had no effect on forage yield regardless of N during the first year. However, a yield response to S by AF-4 tall fescue (Festuca arundinacea, Schreb.) was observed at the highest N rate in the latter part of the second year.

Nutrient Composition and Digestibility. Sulphur has a profound effect on protein synthesis in grass as well as legumes (Oke, 1969; Tisdale, 1977). Sulphur is a component of cysteine and methionine which are two important amino acids in the synthesis of high quality protein. In addition, S fertilization has been reported to lower the non-protein nitrogen and nitrate contents in N fertilized grasses (Cowling and Jones, 1971; Baker et al., 1973).

Bladsoe and Blaser (1947) reported that legumes, in general, have a low protein content when grown on S deficient soils. Similar conclusions were made by Cairns and Carson (1961), who found that S treatment increased the N content and yield of N of alfalfa grown in S-deficient soils. They speculated that the primary effect of S was on N assimilation and other physiological functions of the S containing compounds within the plant. Lancaster et al.

(1971) also observed an increase in the protein content of an orchardgrass and legume sward with S fertilization.

The effect of S fertilization in increasing the S content of forage is well documented (Lancaster et al., 1971; Sheard, 1976; Goh and Kee, 1978; Burmester et al., 1981; Gilbert and Shaw, 1981). Burmester et al. (1981) reported that S uptake was also increased with increasing levels of N, the greatest uptake being at the highest N rate. They concluded that total S uptake by all grasses was directly proportional to the N application rate. Lancaster et al. (1971) showed that fiber and lignin contents were not significantly influenced by S fertilization in the first year.

Supplementation of S has been reported to increase in vitro cellulose digestion (Evans and Davis, 1966; Barton et al., 1971; Bull, 1971). Significant increases in cellulose digestion due to increasing levels of S in the dry matter of the substrate have also been demonstrated by several workers (Starks et al., 1953; Martin et al., 1964; Salsbury and Haenlein, 1964; Evans and Davis, 1966; Barton et al., 1971). Barton et al. (1971) reported the beneficial effect of S upon the in vitro digestibility of lignocellulose in corn (Zea mays) fodder pellets.

Some authors have reported that S has no direct effect on in vivo digestibility of plants (Lancaster et al., 1971;

Goh and Kee, 1978), while, others (Evans and Davis, 1966; Rees et al., 1974) have observed an improvement in dry matter digestibility. Rees et al. (1974), found that S fertilizers increased the dry matter digestibility from 55.2 to 60.2%, reduced the retention time in the reticulo-rumen by 16% and increased the extent of dry matter digestion before the duodenum from 13 to 34% in sheep. They concluded that S fertilizer leads to large increases in both voluntary intake and digestibility of S deficient grass and that fertilizing the grass results in greater improvement than can be achieved by feeding a S supplement.

Animal Parameters. The S content of the diet of the grazing animal would vary, depending on the plant species and plant part consumed, stage of maturity, S status of the soil and season. With low S diets added S increases microbial growth, thereby improving protein synthesis, rate of digestion and dry matter digestibility; all these improvements combined result in an increased voluntary intake (Tisdale, 1977).

If S is deficient an increase in the dietary intake of S by sheep increases both wool and meat production and wool quality (Tisdale, 1977). These improvements were attributed to increased feed intake, increased dry matter, fiber and cellulose digestibility and improved N balance. Odynets et al. (1972) reported that increasing the S content in lamb diets from .1% to .27% resulted in a 10 to 20% increase in wool yield and strength.

In a series of experiments, Bouchard and Conrad (1973) found that S supplemented as sodium sulphate and hydroxy analog of methionine resulted in an increase in dry matter digestibility and milk production in dairy cows. Kennedy and Siebert (1972) showed that the addition of S alone to the diet increased the digestibility and intake of the feed by sheep but had no effect on the digestibility of feed in cattle. However, benefits from S supplementation have been reported in ruminants fed urea-containing diets. Supplementation of S significantly increased the N balance, weight gain and dry matter digestibility in sheep and cattle when supplemental N was provided as urea (Bird, 1974). However, in the absence of adequate N, supplementation of S may be detrimental (Bird, 1974). Bray and Hemsley (1969) observed that when S was added to urea supplemented roughages, the digestibility of the diet, and N and S balances were improved in sheep. Kennedy and Siebert (1972) reported an increase in feed intake, reduction in rumen ammonia concentration and an improvement in N retention when diets supplemented with N and S were fed to cattle.

Lancaster et al. (1971) found that microbial activity as measured by in vitro gas production, increased in grasses and decreased in legumes as S application in the form of sodium sulphate was increased from 0 to 40 ppm. Jones et al. (1970) stated that the gas production in vitro was greatly increased as the S level of the subterraneum clover substrate was increased. Whanger and Matrone (1965) observed that S supplements added to S deficient diets given to sheep increased the concentration of rumen propionic, butyric and higher volatile fatty acids, and decreased rumen level of lactic acid. Hume and Bird (1970) found that raising the S intake from .61 g/d to 1.95 g/d increased the microbial protein synthesis from 82 g/d to 94 g/d in sheep. Brey and Hemsley (1969) reported that sulphate supplements to the diet increased crude fiber digestion, and N and S retention in sheep. They found that the blood urea N levels were decreased with sulphate supplementation. A linear decrease in the concentration of plasma urea N with increasing sulphate supplementation was reported by Kennedy and Siebert (1972). Bray and Hemsley (1969) demonstrated that the addition of sulphates to the diet increased rumen ammonia-nitrogen, but the pH became more acidic.

Nitrogen-to-Sulphur Ratio

The importance of N-to-S ratio in plant and animal nutrition is well documented (Dijkshoorn et al., 1960; O'Connor and Vartha, 1962; Dijkshoorn and Van Wijk, 1967; Kennedy and Siebert, 1972; Metson, 1973; Bird, 1974; Tisdale, 1977). Metson (1973) reported that in conditions where S is in excess or N is deficient, non-protein S may accumulate in the plant tissue, whereas, when S is deficient non-protein nitrogen may accumulate. Under these situations, these forages can cause nutritional disorders in ruminants. According to Metson (1973) the optimum range for N-to-S ratio is between 14:1 to 16:1 for grasses and 10:1 for legumes. Ruminants perform satisfactorily when it is between 10:1 to 12:1 (Tisdale, 1977). Thus, S levels that are adequate for plant growth may be deficient for animal performance.

Effect of Red Clover (*Trifolium pratense*, L.) on Forages

Effect On Dry Matter Yields. The importance of growing grasses in association with legumes has long been recognized as an essential component for increasing pasture productivity in areas where N is very deficient. Beneficial effects of grass-legume mixtures have been repeatedly demonstrated (Cowling, 1961; Carter and Scholl, 1962; Paetzold, 1966; Blaser et al, 1969; Sau, 1970; Hunt et al, 1975; Johansen and Kerridge, 1979). Legumes can be used successfully to meet the N requirements of grasses as well as to improve the yield and quality of grass-legume mixtures (Cowling, 1961; Carter and Scholl, 1962). Beneficial effects of legumes as components in forage mixtures is due effects of legumes as components in forage mixtures is due to the ability of symbiotic Rhizobium bacteria to fix atmospheric N for growth and improved protein content. This symbiotic root interrelationship between legumes and grasses is due to differences in nutritive requirements or habits of root growth of the associated plants which permit more efficient use of the soil, or to excretion of N from nodules of the leguminous plant.

Fertilizer N can be of either organic or inorganic form when applied to the soil. Legume N is fixed by Rhizobium bacteria in organic forms. According to Mulder (1952) these organic compounds may be decomposed by microorganisms and converted to ammonia, nitrate or absorbed by plant roots and fixed in bacterial proteins. These bacterial proteins may be temporarily unavailable to plants. Furthermore, legume N reacts gradually and more evenly than fertilizer N (Mulder, 1952).

Fertilizer N can be lost from soils as ammonia, nitrogen (N_2 gas), or nitrous oxide by biological denitrification or leaching. Nitrogen losses from legume nodules, however, are negligible (Allison, 1966). In addition, fertilizer N may change the soil pH and the uptake of other nutrients.

The N contributions of the clover component on a mixed sward has been estimated by several workers. Wagner (1954)

Holmes and Maclusky (1955) and Cowling (1961) reported it to be about 136 and 184 kg N/ha, respectively. Hunt et al. (1975) showed that the potential dry matter productivity of red clover (Trifolium pratense, L.) is equivalent to grass given fertilizer N at 252, 209 and 144 kg/ha for the first, second and third years respectively. Muller (1975) and Lyubinetskll et al. (1975) estimated the fixation of N by red clver to be 50 to 220 and 88 kg/ha, respectively.

The direct transfer of N from the legume to the associate grass appears to be relatively unimportant. Sears (1961) stated that characteristically red clover does not pass N directly to the associated grasses but only through the grazing animal. Additional N may be, however made available by plowing and decomposition of legume tissues.

Nutrient Composition and Digestibility. Legumes improve the quality of herbage through increasing the crude protein content of the grasses (Birch et al., 1967; Dubbs, 1971; Chestnut, 1974; Haystead and Lowe, 1977). Wagner (1954) reported that the mixed swards produced more total protein than the grasses in pure stand fertilized with as much as 179 kg N/ha. The pasture production was also found to be uniformly distributed throughout the year. Milford (1960) showed that the legumes had higher digestible crude protein contents than the grasses studied. Legumes are highly digestible and rich in N, potassium, Ca, Mg and P

(Visser and Preller, 1965; Appadurai, 1968; Frame et al., 1974).

Animal Parameters. Milford and Minson (1966), and Weston and Hogan (1971) showed that voluntary intake of legume was higher than that of grasses. Similar observations were made by Thornton and Minson (1973) who found the voluntary intake of legumes to be about 28% higher than that of equally digestible grasses. In trials with sheep, voluntary intake of red clover was found to be greater than that of orchardgrass (Prigge, 1974). Blaser et al. (1969) showed that N fertilization did not have any effect on palatability of grass. However, inclusion of the clover component improved the palatability as compared with grass fertilized with N.

YIELD AND NUTRITIONAL VALUE OF ORCHARDGRASS AS INFLUENCED BY NITROGEN AND SULPHUR FERTILIZATION AND ASSOCIATED RED CLOVER

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Summary

The effects of red clover (Trifolium pratense, L.), three levels of nitrogen (N) (0, 224 and 448 kg• ha^{-1}•yr^{-1}, as a split application) and two levels of sulphur (S) (0 and 34 kg•ha^{-1}•yr^{-1}) on yield, chemical composition and dry matter digestibility of an orchardgrass (Dactylis glomerata, L.) sward were studied. The treatments were replicated three times in a randomized block design with a split plot arrangement of treatments. Crude protein content, in vitro dry matter digestibility and dry matter yield were increased ($P<.01$) with increasing levels of N. Increasing levels of N decreased ($P<.05$) S content of orchardgrass. The inclusion of red clover did not have any significant effect on the components analyzed, probably due to a low proportion of red clover (approximately 8%) in the mixed sward. Application of S increased ($P<.05$) the S content of orchardgrass and decreased N-to-S ratio at all levels of N application.

Application of S increased (P<.05) the crude protein and in vitro dry matter digestibility, whereas, neutral detergent fiber, lignin and hemicellulose were decreased (P<.05). Nitrogen fertilization had a quadratic effect (P<.05) on in vivo digestibility of dry matter, acid detergent fiber and hemicellulose and a linear effect (P<.001) on apparent digestibility of crude protein. Inclusion of red clover increased (P<.01) in vivo digestibility of dry matter and hemicellulose, and decreased (P<.01) digestibility of acid detergent fiber and lignin. Application of S had no direct effect on in vivo digestibility of orchardgrass. A linear interaction (P<.01) between N and S was observed for dry matter and crude protein digestibility. Rumen fluid ammonia-N was increased (P<.05) and butyric acid concentration was decreased (P<.05) linearly by N fertilization.

(Key words: Forage Quality, Nitrogen Fertilization, Sulphur Fertilization, Orchardgrass, Sheep)

INTRODUCTION

Sulphur is an essential element for both plants and animals and is a unique nutrient in that large quantities are added to soils from S dioxide through rainfall (Jones et al., 1979). Low analysis, S containing fertilizers have been a significant source of S in the past, but are no longer in common use, reducing this source of supply (Tisdale and Nelson, 1975). Studies on forages have shown that responses to heavy N fertilization were often limited by a deficiency of S (Stewart and Porter, 1967; Aulakh et al., 1976). An insufficient supply of S can affect both the yield and quality of the herbage produced (Cairns et al., 1961; Sorenson et al., 1968). Under conditions of high N fertilization and S deficiency, protein synthesis is reduced by a lack of the S containing amino acids, cysteine and methionine (Dijkshoorn et al., 1960), and this is reflected by marked accumulation of N in the plant as nitrate-nitrogen, amides and free amino acids (Terman et al., 1976; Goh and Kee, 1978). Such herbages are nutritionally unbalanced and can cause nutritional disorders in ruminants. Thus, the ratio of total N to total S becomes important as a measure of protein production and quality of the forage (Tisdale, 1977).

Although the effects of N and S on the dry matter yield of forages are well documented, information on their effects on the nutrient composition of herbage is lacking. Therefore, the objectives of the present study were to: investigate the effects of N and S fertilization and the N contribution of red clover on the dry matter yields, nutrient composition and in vitro dry matter digestibility of an orchardgrass sward; and to compare the effects of these treatments on in vivo nutrient digestibility, N balance, and rumen and blood parameters of sheep.

EXPERIMENTAL PROCEDURE

The forages used were grown at the Shenandoah Valley Research Station, Steeles Tavern, Virginia in a Fredrick-Cherty loam soil with a pH of 6.8. An established orchardgrass sward was used. Plots measuring 36x43 m^2 were arranged in a randomized block design with a split-plot arrangement of treatments with three replications (figure 1). The treatments on the orchardgrass with and without 33.6 kg S/ha were:

None;

224 kg N $ha^{-1} \bullet yr^{-1}$;

448 kg N $ha^{-1} \bullet yr^{-1}$;

Red clover;

Phosphorous (P), potassium (K) and magnesium (Mg) (112 kg P/ha, 112 kg K/ha, 24 kg Mg/ha as chelates combined with 33.6 kg N/ha in a liquid) were applied uniformly to the entire area, according to the soil test. Red clover (cultivar-Kenstar) was seeded at the rate of 8.96 kg/ha using a cultipack seeder in September, 1980 and March, 1981.

Three levels of nitrogen (N), 0, 224 and 448 $kg \cdot ha^{-1} \cdot yr^{-1}$ were applied as NH_4NO_3 in two split applications (March 31, 1981, and July 2, 1981) to the pure stand. Sulphur (S) was applied at the rate of 0 and 33.6 kg ha^{-1} yr^{-1} as $CaSO_4$ to one half of each plot.

Harvesting and Sampling of Forages

Plots were harvested on August 4, 1981. Each replicate was divided into three strata, and three quadrats of 60x60 cm^2 were taken from each stratum. The plots for treatments were clipped to a height of 7 cm. Herbage from quadrats within each stratum was then composited, and grass and legume were separated in the mixed sward to determine percent of clover. Representative samples were taken to determine the dry matter content by oven drying the samples at 60 C for 24 hr and used in subsequent laboratory analysis. All plots were harvested separately, using a mower-conditioner and field dried before baling. Weight of hay bales were taken for each replicate in each treatment separately and used to calculate the yield per hectare.

Chemical Analysis

All analyses were made on oven-dried orchardgrass

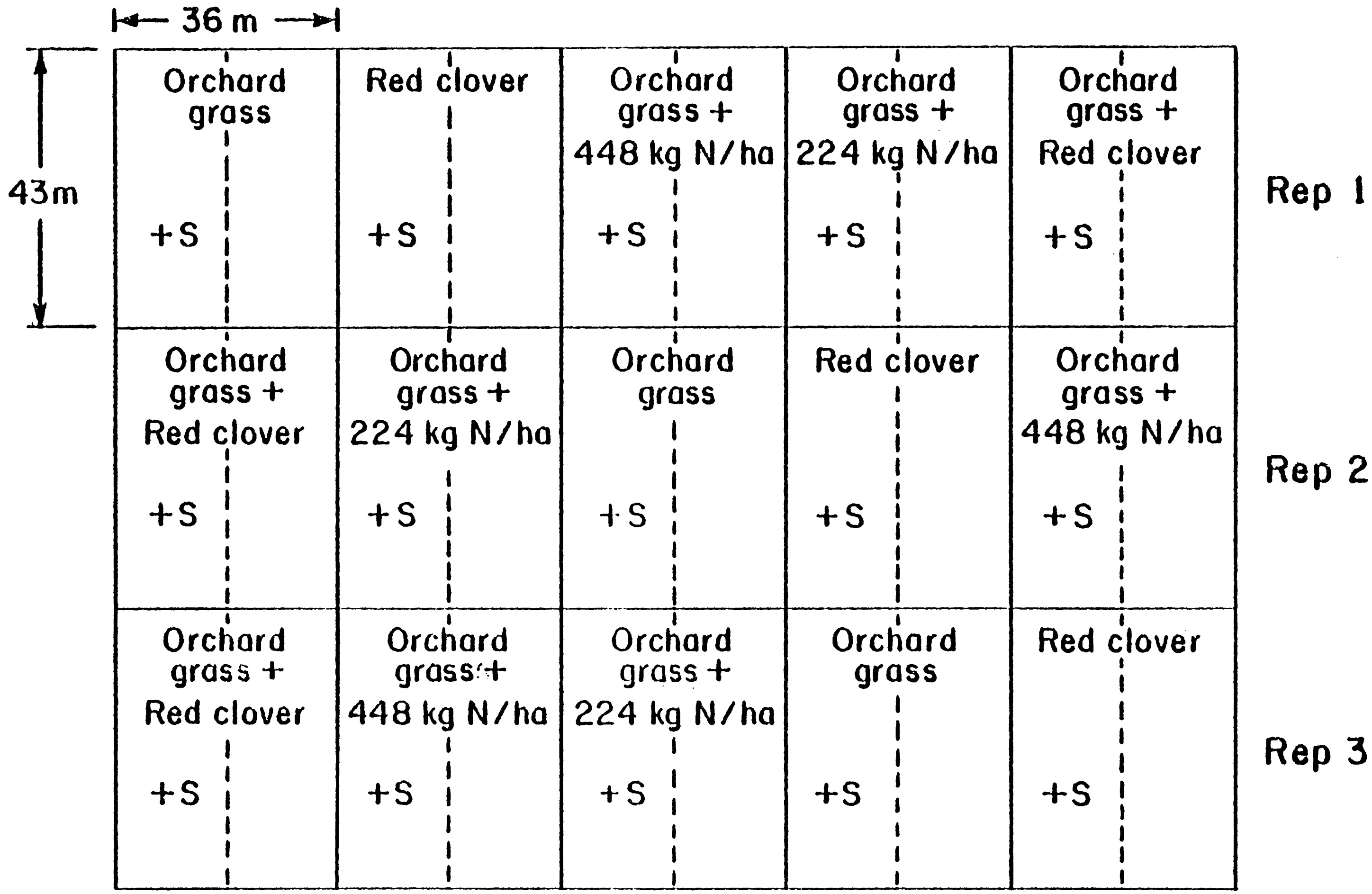

Figure 1. Design: Experimental Plots

samples, ground to pass 1 mm sieve. The samples were analyzed for dry matter, N by the Kjeldhal method (A.O.A.C., 1980), neutral detergent fiber (Van Soest and Wine, 1967), acid detergent fiber (Van Soest, 1963), lignin and cellulose (Van Soest and Wine, 1968). In vitro dry matter digestibility was determined according to the method of Tilley and Terry (1963) modified by Barnes (1969) and using a buffer solution described by McDougall (1948).

One-half gram samples were wet ashed with a 3:1 nitric-perchloric acid for mineral analysis and lanthanum oxide was included in the dilution for calcium (Ca) and magnesium (Mg) analysis. Total S in the plant tissue was determined colorimetrically (Tabatabi et al., 1970). Calcium and Mg were determined by atomic absorption spectrophotometer.

Metabolism Trial

Three metabolism trials were conducted each involving 16 wether lambs (35 kg). The animals were placed in two blocks of eight animals each by weight. Sheep within each block were allotted at random to the eight forages described previously. The herbage was fed in the form of chopped (3.8 cm) hay.

The sheep were treated for internal parasites and placed in false bottom metabolism stalls similar to those of Briggs and Gallup (1949), permitting separate collection of urine and feces. There was a 2-wk adaptation period to the stalls followed by a 3-d period during which animals were fed orchardgrass hay. The lambs were fed 800 g of chopped orchardgrass hay and 10 g of salt daily. One-half of the daily allowance was fed at 0700 h and the other half at 1900 h. Water was available at all times except during the 2-h feeding periods. A 7-d preliminary period preceded a 7-d period during which total feces and urine were collected.

The diets were sampled at each feeding from 2 d before the start, until 2 d prior to the end of the 7-d collection period. The samples were composited at the end of the trial. At the end of the collection period the refusals from each animal were weighed and subsampled. Feces were collected daily, dried for 24 h at 60 C in a forced-draft oven, allowed to air-equilibrate, and composited by animal in metal cans with loose fitting lids. At the end of the collection period, the feces from each animal were weighed, mixed and subsampled.

Urine was collected in plastic jugs containing 15 ml of a 1:1 (w/w) solution of concentrated sulphuric acid and water and approximately 500 ml of water. The urine was collected every 24 h, brought to a constant weight with water and a 2% sample by volume was placed in an airtight plastic bottle. Daily samples were composited by animal and refrigerated. The urine samples were subsampled at the end of the collection period and frozen for subsequent analysis.

Rumen ingesta samples were taken via stomach tube 2 h after the morning feeding at the begining and end of the collection period in each trial. The rumen fluid was strained through four layers of cheesecloth for the determination of pH (pH meter), ammonia N (Conway, 1958) and volatile fatty acids (Erwin et al., 1961). Volatile fatty acid levels were determined with a Perkin-Elmer 881 gas chromatograph. Blood samples, taken by jugular puncture on the same day 6 h after feeding, were analyzed for blood urea N (Coulombe and Favreau, 1963) and serum Ca and Mg, using an atomic absorption spectrophotometer.

The feed, fecal and refusal samples were analyzed as described for analysis of forage samples. Kjeldahl N (A.O.A.C., 1980) was determined on duplicate 5 ml urine samples.

Data were statistically analyzed using a general linear model of Barr et al. (1976). Six contrasts were made to partition the treatment variance of significance using SAS package programs.

RESULTS AND DISCUSSION

Effects on Dry Matter and Crude Protein Yields

Dry matter yield of orchardgrass was increased almost fourfold with 224 kg N/ha, but was not increased further by applying 448 kg N/ha. This represents a quadractic response

(P<.01). Crude protein yield was increased (P<.001) linearly with the application of N fertilizer (table 1). However, the difference between application rates of 224 and 448 kg N/ha was small. This increase in yield with N fertilization is in general agreement with other published reports (Noller and Rhykerd, 1974; Donohue, 1975; Sauerbery, 1975; Kunelius et al., 1977). It is well established that increasing the supply of N increases the number of tillers per plant, size of the leaf and shoot/root ratio, thereby, increasing the yield (Whitehead, 1970). Crude protein and dry matter yields from the total of grass-clover mixture were similar to that of pure orchardgrass not receieving fertilizer N, indicating little or no underground transfer of fixed N from clover to the companion grass. Butler and Bethurst (1956) reported that this transfer occurs principally through the sloughing off and decay of nodules. The low proportion of red clover (approximately 8%) in the sward also may have contributed to this non significant effect.

Although a trend towards an increase in dry matter and crude protein yields were observed with S fertilization, the differences were not statistically significant (table 2). Burmester et al. (1981) reported that S had no effect on forage yield during the first year.

TABLE 1. DRY MATTER AND CRUDE PROTEIN YIELDS[a] OF ORCHARDGRASS FERTILIZED WITH DIFFERENT NITROGEN LEVELS OR GROWN IN ASSOCIATION WITH RED CLOVER

Component	Treatments				
	N fertilization, kg/ha			Red clover	SEM
	0	224	448		
	kg/ha				
Dry matter[b]	652	2359	2302	742	104
Crude protein[c]	79	374	404	92	17

[a] Dry matter basis.

[b] Quadratic effect of N fertilization (P<.001).

[c] Linear effect of N fertilization (P<.001).

TABLE 2. EFFECT OF SULPHUR FERTILIZATION ON DRY MATTER AND CRUDE PROTEIN[a] YIELDS OF ORCHARDGRASS.

Component	S fertilization, kg/ha		SEM
	0	34	
	----------kg/ha----------		
Dry matter	1496	1531	73
Crude protein	224	250	12

[a] Dry matter basis.

<u>Effect on Nutrient Composition</u>

Nitrogen fertilization resulted in a linear increase ($P<.01$) in the crude protein content of herbage (table 3). It is well known that increases in the rate of fertilizer N to grass results in an increase in the N content of the herbage as protein-N or non protein-N and result in a progressive increase in the total N content of the herbage (Reid, 1966; Rhykerd et al., 1966; Cowling et al., 1967; Lampeter et al., 1973; Cucu et al., 1973; Noller et al., 1974; Hojjati et al., 1977). The mean crude protein content of companion grass in the orchardgrass-clover mixture was similar to that of grass grown alone without N fertilizer (table 3), probably due to the low proportion of legume in the sward.

The application of 224 kg N/ha increased neutral detergent fiber (NDF) and hemicellulose but application of 448 kg N/ha reduced these to approximately the same levels as the unfertilized grass (quadratic effect, $P<.01$). A linear decrease ($P<.01$) in acid detergent fiber and cellulose contents was observed (table 3). On the contrary, Reid et al. (1966) reported that N had little effect on cellulose, acid detergent fiber, neutral detergent fiber and lignin of orchardgrass hay. However, Reid et al. (1967) found a small reduction in the cellulose and acid-insoluble lignin content of orchardgrass with levels of fertilizer N

TABLE 3. CRUDE PROTEIN, FIBER COMPONENTS[a] AND IN VITRO DRY MATTER DIGESTIBILITY OF ORCHARDGRASS FERTILIZED WITH DIFFERENT NITROGEN LEVELS OR GROWN IN ASSOCIATION WITH RED CLOVER

Component	Treatments				
	N fertilization, kg/ha			Red clover	SEM
	0	224	448		
	---------------	%	---------------		
Crude protein[b]	12.2	15.9	17.6	12.3	.4
Neutral detergent fiber[c]	65.8	67.6	64.6	66.1	.5
Acid detergent fiber[b]	37.2	36.7	35.3	37.2	.3
Lignin[b]	5.5	6.6	6.4	5.3	.1
Cellulose[b]	30.4	29.3	28.5	31.5	.3
Hemicellulose[c]	28.6	31.0	29.4	28.9	.4
In vitro dry matter digestibility[b]	54.4	56.4	58.2	55.4	.4

[a] Dry matter basis.

[b] Linear effect of N fertilization (P<.01).

[c] Quadratic effect of N fertilization (P<.01).

up to 504 kg/ha. A linear increase in lignin content and in vitro dry matter digestibility ($P<.01$) was observed with N fertilization (table 3). Similar trends have been reported by others (Reid et al., 1966; Belasko, 1977; Goh and Kee, 1978). Inclusion of red clover had no effect on the composition and in vitro digestibility of the associated grass (table 3).

Application of S increased ($P<.05$) the crude protein content and decreased ($P<.05$) the neutral detergent fiber, lignin and hemicellulose contents (table 4). The increase in crude protein content is probably associated with S being a component of cystine and methionine which are two important amino acids for the synthesis of high quality protein. Oke (1969) and Tisdale (1977) reported that S has a profound effect on protein synthesis in grass as well as in the legumes. On the contrary, Lancaster et al. (1971) showed that fiber and lignin percentage were not significantly influenced by S fertilization in the first year. Sulphur had no effect on acid detergent fiber and cellulose contents (table 4).

Published information on the effects of S on in vitro dry matter digestibility is conflicting. Lancaster et al. (1971), and Goh and Kee (1978) reported that S had no direct effect on the digestibility of plants. However, Evans and Davis (1966) reported that addition of sulphate to an in

TABLE 4. EFFECT OF SULPHUR FERTILIZATION ON THE CRUDE PROTEIN, FIBER COMPONENTS[a] AND IN VITRO DRY MATTER DIGESTIBILITY OF ORCHARDGRASS

Component	S fertilization, kg/ha		SEM
	0	34	
	-----------%-----------		
Crude protein[b]	14.0	15.0	.3
Neutral detergent fiber[b]	66.6	65.5	.3
Acid detergent fiber	36.4	36.8	.2
Lignin[b]	6.1	5.8	.1
Cellulose	29.7	30.2	.2
Hemicellulose[b]	30.2	28.7	.3
In vitro dry matter digestibility[b]	55.1	57.2	.3

[a]Dry matter basis.

[b]Different at (P<.05).

vitro system improved cellulose digestibility. In the present study, S increased ($P<.05$) the in vitro dry matter digestibility of orchardgrass from 55.1 to 57.2%.

Nitrogen fertilization at 224 kg N/ha decreased Mg and Ca contents of orchardgrass (table 5), and application of 448 kg N/ha increased the contents above the control values (quadratic effect, $P<.01$). Reid et al. (1964) and Hojjati et al. (1977) reported that N fertilization increased the Mg content whereas Reid et al. (1966) found no consistent effect of N fertilization on Ca content. Increasing levels of N decreased ($P<.001$) the S content of orchardgrass and increased ($P<.001$) the N-to-S ratio of the sward linearly. The effect of N fertilization on decreasing S content is in agreement with the results of O'Connor and Vartha (1969). However, others (Jones, 1960; Lancaster et al., 1971; Burmester et al., 1981) have reported an increase in S content with increasing levels of N application.

Sulphur fertilization increased ($P<.05$) the Mg and Ca content of orchardgrass (table 6). It also increased ($P<.05$) the S content of orchardgrass and decreased ($P<.05$) N-to-S ratio averaged over all N levels. This increase in Mg and Ca is probably due to the presence of high levels of sulphate in the soil.

Interaction between N and S has been reported by O'Connor et al. (1969) and Goh and Kee (1978). However, in

TABLE 5. MAGNESIUM, CALCIUM, SULPHUR CONTENTS AND NITROGEN-TO-SULPHUR RATIO OF ORCHARDGRASS[a] FERTILIZED WITH DIFFERENT NITROGEN LEVELS OR GROWN IN ASSOCIATION WITH RED CLOVER

ITEM	Treatments				
	N fertilization, kg/ha			Red clover	SEM
	0	224	448		
Magnesium, %[b]	.23	.21	.28	.22	.01
Calcium, %[b]	.50	.45	.52	.48	.01
Sulphur, %[c]	.27	.21	.18	.24	.01
Nitrogen:sulphur[c]	7.8	12.6	16.7	8.8	.47

[a]Dry matter basis.

[b]Quadratic effect of N fertilization ($P<.001$).

[c]Linear effect of N fertilization ($P<.001$).

TABLE 6. EFFECT OF SULPHUR FERTILIZATION ON THE MAGNESIUM, CALCIUM, SULPHUR CONTENTS AND NITROGEN-TO-SULPHUR RATIO OF ORCHARDGRASS[a]

ITEM	S fertilization, kg/ha		SEM
	0	34	
Magnesium, %[b]	.23	.24	.01
Calcium, %[b]	.47	.51	.01
Sulphur, %[b]	.17	.28	.01
Nitrogen:sulphur[b]	13.6	9.3	.33

[a]Dry matter basis

[b]Different at ($P<.05$)

the present study, no such interactions were observed for any of the parameters studied (tables 7,8 and 9). However, there was a trend for S fertilization to increase crude protein more at the high levels of N fertilization (table 7). Decreases in the N-to-S ratios with S application were observed at all levels of N fertilization.

Effects on In Vivo Digestibility and Nitrogen Retention

The composition of the forages is shown in table 10. Nitrogen fertilization at 224 kg N/ha increased ($P<.05$) in vivo dry matter digestibility of orchardgrass hay (table 11). The values for forage fertilized with 448 kg N/ha were intermediate, indicating maximum response was reached at the medium level of fertilization (quadratic effect, $P<.05$). Several authors have reported that N fertilization increased the in vitro dry matter digestibility (Reid et al., 1966; Belasko, 1977; Goh and Kee, 1977) by delaying the herbage maturity. Application of N to orchardgrass resulted in a linear increase ($P<.05$) in digestibility of crude protein (table 11), probably a result of increased crude protein content of the forage. Poulton et al. (1957) observed that crude protein digestibility was increased as N fertilization was increased from 112 to 224 kg/ha. Bratzler et al. (1959) reported that N fertilization up to 336 kg/ha increased crude protein digestibility.

TABLE 7. DRY MATTER AND CRUDE PROTEIN[a] YIELDS OF ORCHARDGRASS FERTILIZED WITH DIFFERENT NITROGEN LEVELS OR GROWN IN ASSOCIATION WITH RED CLOVER AS AFFECTED BY SULPHUR FERTILIZATION

		Treatments				
Component	Sulphur fertilization, kg/ha	N fertilization, kg/ha 0	224	448	Red clover	SEM
		----------kg/ha----------				
Dry matter	0	659	2262	2362	701	146
	34	644	2455	2243	782	146
Crude protein	0	78	336	398	86	24
	34	80	411	410	98	24

[a] Dry matter basis.

TABLE 8. CRUDE PROTEIN, FIBER[a] AND IN VITRO DRY MATTER DIGESTIBILITY OF ORCHARDGRASS FERTILIZED WITH DIFFERENT NITROGEN LEVELS OR GROWN IN ASSOCIATION WITH RED CLOVER AS AFFECTED BY SULPHUR FERTILIZATION

Component	Sulphur fertilization, kg/ha	Treatments				SEM
		N fertilization, kg/ha			Red clover	
		0	224	448		
		%				
Crude protein	0	11.9	14.9	17.0	12.2	.6
	34	12.5	16.8	18.3	12.4	.6
Neutral detergent fiber	0	66.5	68.3	65.4	66.1	.6
	34	65.1	66.9	63.9	66.1	.6
Acid detergent fiber	0	37.7	36.3	34.6	37.0	.4
	34	36.7	37.0	35.9	37.4	.4
Lignin	0	5.6	6.9	6.5	5.4	.2
	34	5.3	6.4	6.3	5.1	.2
Cellulose	0	30.7	28.5	28.5	31.1	.5
	34	30.2	30.1	28.6	31.9	.5
Hemicellulose	0	28.9	32.0	30.8	29.2	.6
	34	28.3	30.0	27.9	28.6	.6
In vitro dry matter digestibility	0	53.4	54.8	57.2	54.8	.6
	34	55.4	58.0	59.1	56.0	.6

[a] Dry matter basis

TABLE 9. MAGNESIUM, CALCIUM, SULPHUR CONTENTS AND NITROGEN-TO-SULPHUR RATIO OF ORCHARDGRASS[a] FERTILIZED WITH DIFFERENT NITROGEN LEVELS OR GROWN IN ASSOCIATION WITH RED CLOVER AS AFFECTED BY SULPHUR FERTILIZATION

ITEM	Sulphur fertilization, kg/ha	Treatments: N fertilization, kg/ha 0	224	448	Red clover	SEM
Magnesium, %	0	.21	.22	.26	.22	.01
	34	.24	.21	.29	.22	.01
Calcium, %	0	.48	.46	.49	.47	.02
	34	.53	.45	.56	.48	.02
Sulphur, %	0	.20	.17	.14	.18	.01
	34	.33	.26	.21	.31	.01
Nitrogen:sulphur	0	9.4	14.5	19.6	11.0	.67
	34	6.2	10.7	13.8	6.5	.67

[a] Dry matter basis

TABLE 10. CHEMICAL COMPOSITION OF ORCHARDGRASS FERTILIZED WITH DIFFERENT NITROGEN LEVELS AND AN ORCHARDGRASS-RED CLOVER MIXTURE AS AFFECTED BY SULPHUR FERTILIZATION

		Treatments				
Component	Sulphur fertilization, kg/ha	N fertilization, kg/ha			Red clover	SEM
		0	224	448		
Dry matter, %	0	92.2	91.9	92.4	91.7	.44
	34	92.6	91.7	92.2	92.7	.44
Composition of dry matter, %						
Crude protein [a]	0	13.8	16.2	17.4	14.6	.40
	34	15.0	16.6	18.0	13.8	.40
Neutral detergent fiber [b]	0	58.0	62.3	61.0	56.6	1.07
	34	58.6	65.6	59.7	57.9	1.07
Acid detergent fiber [b]	0	35.9	37.0	36.2	35.6	.75
	34	35.5	39.7	35.1	35.5	.75
Cellulose [b]	0	29.0	29.9	29.5	28.4	.58
	34	28.4	31.8	28.3	28.7	.58
Hemicellulose [b]	0	22.2	25.2	24.8	20.9	.49
	34	23.1	26.0	24.5	22.4	.49
Lignin [b]	0	5.3	6.0	5.6	6.2	.25
	34	5.8	6.6	5.5	5.7	.25

[a] Linear effect of N fertilization (P<.001).

[b] Quadratic effect of N fertilization (P<.05).

TABLE 11. IN VIVO DIGESTIBILITY OF ORCHARDGRASS FERTILIZED WITH DIFFERENT LEVELS OF NITROGEN AND AN ORCHARDGRASS-RED CLOVER MIXTURE

Component	Treatments				
	N fertilization, kg/ha			Red clover	SEM
	0	224	448		
	%				
Dry matter[a,b]	57.3	61.0	59.1	59.9	.6
Crude protein[c]	67.6	68.8	71.2	68.1	.6
Neutral detergent fiber	59.3	61.3	58.4	58.6	.7
Acid detergent fiber[a,b]	55.6	57.3	53.9	51.6	.9
Lignin[b,c]	37.2	37.8	28.0	25.6	1.9
Cellulose	59.1	62.0	58.1	56.7	1.0
Hemicellulose[a,b]	64.7	67.4	64.7	70.2	.8

[a]Quadratic effect of N fertilization ($P<.05$).

[b]Orchardgrass vs red clover ($P<.05$).

[c]Linear effect of N fertilization ($P<.05$).

Application of N did not have any significant effect on the digestibility of neutral detergent fiber (NDF), and cellulose (table 11). Maximum digestibility of acid detergent fiber (ADF) and hemicellulose was for the forage fertilized with 224 kg N/ha (quadratic effect, P<.05) (table 11). Application of 448 kg N/ha resulted in a decrease in lignin (P<.05) digestibility of orchardgrass. On the other hand, Poulton et al. (1957) and Bratzler et al. (1959), reported that N fertilization had little effect on the digestibility of cellulose, ADF, NDF and lignin of orchardgrass hay.

Fertilizer N had no significant effect on N retention of sheep (table 12). Seidler et al. (1972) reported that N balance was greatest for the orchardgrass hay fertilized with 640 kg N/ha N. The lack of effect of N fertilizer on N retention observed in our study may be attributed to the possible imbalance between total N and soluble carbohydrate contents in the orchardgrass hay, which may have limited its use by rumen microorganisms.

Inclusion of red clover in orchardgrass increased (P<.05) the in vivo digestibility of dry matter and hemicellulose, and decreased (P<.05) the digestibility of ADF and lignin (table 11). These effects may have been due to higher digestibility of red clover. Red clover had no significant effect on the digestibility of crude protein and

TABLE 12. NITROGEN BALANCE OF SHEEP FED ORCHARDGRASS FERTILIZED WITH DIFFERENT LEVELS OF NITROGEN AND AN AN ORCHARDGRASS-RED CLOVER MIXTURE

ITEM	Treatments[a]				SEM
	N fertilization, kg/ha			Red clover	
	0	224	448		
Intake, g/d[b]	16.9	18.98	20.70	16.61	.29
Excretion, g/d					
Fecal[b]	5.48	5.92	5.93	5.29	.11
Urinary[b]	10.33	11.26	13.34	9.17	.42
Total[b]	15.81	17.18	19.27	14.46	.42
Retention					
Grams per day	1.09	1.80	1.43	2.15	.54
Percent of intake	6.45	9.48	6.90	12.94	2.92
Percent of absorbed	9.55	13.78	9.70	18.99	4.30

[a]Each value represents the mean of twelve animals.

[b]Linear effect of N fertilization ($P<.01$).

cellulose fractions.

Applications of S had no direct effect on in vivo digestibility and N retention of orchardgrass (tables 13 and 14). However, a linear interaction ($P<.01$) between N and S was observed with regard to the digestibility of dry matter and crude protein (table 15). O'Connor and Vartha (1969) and, Goh and Kee (1978) reported an interaction between N and S on dry matter yields of orchardgrass and perennial ryegrass (Lolium perenne, L.). In their experiments apparent digestibility of dry matter was not affected by N fertilization in plots not fertilized with S, but was related to level of N application when the sward was fertilized with S. In the research reported here digestibility of crude protein was increased by the low level of N fertilization (224 kg/ha), regardless of whether or not S was applied, but a response to 448 kg N/ha was recorded only when S was applied. O'Connor and Vartha (1969), Scattini (1981) and Burmester et al. (1981) reported that the pastures responded to fertilizer containing sulphates at high N rates. These results indicate that response to N fertilization depends on S application to soil if S is limiting. No interaction between N and S was observed on N retention of sheep (table 16).

TABLE 13. EFFECT OF SULPHUR FERTILIZATION ON IN VIVO DIGESTIBILITY OF ORCHARDGRASS AND AN ORCHARDGRASS-RED CLOVER MIXTURE

Component	S fertilization, kg/ha		SEM
	0	34	
	---------%----------		
Dry matter	59.1	59.6	.4
Crude protein	68.6	69.3	.4
Neutral detergent fiber	58.9	59.8	.5
Acid detergent fiber	54.2	55.0	.6
Lignin	31.3	33.0	1.4
Cellulose	59.0	59.1	.7
Hemicellulose	66.4	67.1	.6

TABLE 14. NITROGEN BALANCE OF SHEEP FED SULPHUR FERTILIZED ORCHARDGRASS AND AN ORCHARDGRASS-RED CLOVER MIXTURE [a]

ITEM	S fertilization, kg/ha		SEM
	0	34	
Intake, g/d	18.08	18.51	.20
Excretion, g/d			
Fecal	5.66	5.65	.08
Urinary	11.27	11.34	.30
Total	16.93	16.99	.30
Retention			
Grams per day	1.15	1.52	.38
Percent of intake	6.36	8.21	2.06
Percent of absorbed	9.26	11.82	3.04

[a] Each value represents the mean of 24 animals.

TABLE 15. IN VIVO DIGESTIBILITY OF ORCHARDGRASS FERTILIZED WITH DIFFERENT LEVELS OF NITROGEN AND AN ORCHARDGRASS-RED CLOVER MIXTURE AS AFFECTED BY SULPHUR FERTILIZATION

Component	Sulphur fertilization, kg/ha	Treatments: N fertilization, kg/ha 0	224	448	Red clover	SEM
		------------%------------				
Dry matter[a]	0	58.0	61.6	57.2	59.5	.8
	34	56.6	60.4	61.1	60.3	.8
Crude protein[a]	0	69.1	68.2	69.1	67.8	.8
	34	66.1	69.4	73.3	68.4	.8
Neutral detergent fiber	0	58.5	62.0	56.7	58.4	1.0
	34	60.1	60.5	60.0	58.8	1.0
Acid detergent fiber	0	54.9	58.7	52.3	51.0	1.3
	34	56.3	56.0	55.6	52.2	1.3
Lignin	0	35.1	38.8	24.2	27.0	2.8
	34	39.2	36.7	31.9	24.2	2.8
Cellulose	0	58.4	63.4	57.0	56.8	1.4
	34	59.8	60.7	59.3	56.7	1.4
Hemicellulose	0	63.6	67.5	63.1	71.2	1.1
	34	65.8	67.3	66.2	69.3	1.1

[a] Linear effect of NxS fertilization (P<.01).

TABLE 16. NITROGEN BALANCE OF SHEEP FED ORCHARDGRASS FERTILIZED WITH DIFFERENT LEVELS OF NITROGEN AND AN ORCHARDGRASS-RED CLOVER MIXTURE AS AFFECTED BY SULPHUR FERTILIZATION

ITEM	Sulphur fertilization, kg/ha	Treatments[a] N fertilization, kg/ha 0	224	448	Red clover	SEM
Intake, g/d[b]	0	16.14	18.86	20.31	17.02	.41
	34	17.65	19.09	21.08	16.19	.41
Excretion, g/d						
Fecal[b,c]	0	5.00	5.99	6.23	5.47	.16
	34	5.98	5.84	5.62	5.12	.16
Urinary[b,c]	0	9.51	10.63	14.91	10.02	.60
	34	11.15	11.87	14.03	8.31	.60
Total[b,c]	0	14.51	16.62	21.14	15.49	.60
	34	17.13	17.71	19.66	13.43	.60
Retention						
Grams per day	0	1.63	2.24	-.83	1.53	.76
	34	.52	1.38	1.42	2.76	.76
Percent of intake	0	10.10	11.88	-4.09	8.99	4.13
	34	2.95	7.23	6.74	17.05	4.13
Percent of absorbed	0	14.76	17.41	-5.89	13.25	6.07
	34	4.46	7.79	9.18	14.93	6.07

[a] Each value represents the mean of 6 animals.

[b] Orchardgrass differs from red clover (P<.05).

[c] Linear effect of NxS (P<.05).

Effects on Rumen and Blood Parameters

Effects of red clover and N fertilized orchardgrass on pH, ammonia-N, volatile fatty acids and acetic: propionic ratio in the ruminal fluid are shown in table 17. Nitrogen fertilizer linearly increased ($P<.05$) the pH, ammonia-N, iso-butyric and iso-valeric acid content in the rumen fluid and linearly decreased ($P<.05$) the butyric acid content. There were no significant effects of N fertilization on acetic, propionic, valeric and acetic:propionic ratio in the rumen fluid. The high levels of ammonia-N in the rumen fluid may be explained by the higher N content in N fertilized grasses (Frens, 1960). Inclusion of red clover in orchardgrass increased ($P<.05$) the rumen pH and decreased ($P<.05$) the ammonia-N content.

Nitrogen fertilization linearly increased ($P<.01$) the blood urea-N content of sheep fed orchardgrass, whereas, no significant responses were observed for serum Ca and Mg (table 18). Our results are in agreement with Carver et al. (1978) who reported an increase in plasma urea-N levels of steers with increasing levels of N fertilization to the forage.

Sulphur fertilization had no effect on pH, ammonia-N, volatile fatty acid content and acetic:propionic ratio in the rumen fluid (table 19), but increased ($P<.05$) the blood urea-N level (table 20). To the contrary, Hemsley (1969)

TABLE 17. RUMINAL pH, AMMONIA, VOLATILE FATTY ACIDS AND ACETIC-PROPIONIC RATIO IN SHEEP FED ORCHARDGRASS FERTILIZED WITH DIFFERENT NITROGEN LEVELS AND AN ORCHARDGRASS-RED CLOVER MIXTURE

Components	Treatments				SEM
	N fertilization, kg/ha			Red clover	
	0	224	448		
pH[a,b]	6.84	6.91	7.03	6.97	.04
Ammonia-N, mg/dl[a,b]	23.76	26.89	31.11	19.71	1.32
Volatile fatty acids, moles/100 moles					
Acetic	72.44	73.16	73.42	71.89	2.39
Propionic	16.49	16.40	15.78	16.40	.42
Isobutyric[a]	1.28	1.37	1.60	1.38	.05
Butyric[a]	7.06	6.31	6.13	7.83	.30
Isovaleric[a]	1.47	1.60	1.83	1.56	.06
Valeric	1.26	1.16	1.24	.94	.15
Acetic:propionic	4.43	4.51	4.70	4.39	.14

[a]Linear effect of N fertilization (P<.05).

[b]Orchardgrass vs red clover (P<.05).

TABLE 18. BLOOD UREA NITROGEN CONTENT AND SERUM MINERALS OF SHEEP FED ORCHARDGRASS FERTILIZED WITH DIFFERENT NITROGEN LEVELS AND AN ORCHARDGRASS-RED CLOVER MIXTURE

Component	Treatments				SEM
	N fertilization, kg/ha			Red clover	
	0	224	448		
	mg/dl				
Blood urea nitrogen[a]	24.99	26.61	31.22	24.35	1.40
Serum calcium	1.06	1.08	1.06	1.08	.02
Serum magnesium	2.12	2.27	2.19	2.14	.05

[a]Linear effect of N fertilization (P<.01).

TABLE 19. EFFECT OF SULPHUR FERTILIZATION OF ORCHARDGRASS AND AN ORCHARDGRASS-RED CLOVER MIXTURE ON RUMINAL pH, AMMONIA, VOLATILE FATTY ACIDS AND ACETIC-PROPIONIC RATIO IN THE RUMEN FLUID

Component	S fertilization, kg/ha		SEM
	0	34	
pH	6.93	6.94	.03
Ammonia-N, mg/dl	26.14	24.61	.93
Volatile fatty acids, moles/100 moles			
Acetic	73.20	72.25	1.69
Propionic	15.79	16.76	.29
Isobutyric	1.40	1.42	.04
Butyric	6.77	6.87	.21
Isovaleric	1.63	1.61	.04
Valeric	1.21	1.09	.11
Acetic:propionic	4.68	4.31	.10

TABLE 20. EFFECT OF SULPHUR FERTILIZATION OF ORCHARDGRASS AND AN ORCHARDGRASS-RED CLOVER MIXTURE ON BLOOD UREA NITROGEN CONTENT AND SERUM MINERALS OF SHEEP

Component	S fertilization, kg/ha		SEM
	0	34	
	--------mg/dl--------		
Blood urea nitrogen[a]	25.35	28.24	.99
Serum calcium	1.06	1.08	.01
Serum magnesium	2.21	2.15	.03

[a] Different at (P<.05)

found that the blood urea N levels decreased with S fertilization. Quadratic effects (P<.05) of N and S on iso-butyric and iso-valeric acid content in rumen fluid were also observed (table 21). There was no significant NxS interaction on blood urea N, serum Ca and mangesium of sheep (table 22).

TABLE 21. RUMINAL pH, AMMONIA, VOLATILE FATTY ACIDS AND ACETIC-PROPIONIC RATIO OF SHEEP FED ORCHARDGRASS FERTILIZED WITH DIFFERENT LEVELS OF NITROGEN AND AN ORCHARDGRASS-RED CLOVER MIXTURE AS AFFECTED BY SULPHUR FERTILIZATION

Component	Sulphur fertilization, kg/ha	Treatments: N fertilization, kg/ha 0	224	448	Red clover	SEM
pH	0	6.80	6.99	7.05	6.89	.05
	34	6.87	6.84	7.00	7.06	.05
Ammonia-N, mg/dl	0	4.26	27.23	31.34	21.72	1.87
	34	3.26	26.56	30.88	17.75	1.87
Volatile fatty acids moles/100 moles						
Acetic	0	72.31	71.31	75.38	73.85	3.38
	34	72.56	75.01	71.46	63.56	3.38
Propionic	0	16.21	17.08	15.04	14.82	.59
	34	16.77	15.72	16.51	16.55	.59
Isobutyric[a]	0	1.25	1.57	1.53	1.23	.08
	34	1.31	1.18	1.68	1.40	.08
Butyric	0	7.25	6.71	5.29	7.84	.42
	34	6.88	5.91	6.96	7.12	.42
Isovaleric[a]	0	1.47	1.83	1.80	1.41	.08
	34	1.48	1.39	1.86	1.56	.08
Valeric	0	1.53	1.52	.95	.84	.21
	34	.99	.81	1.53	.96	.21
Acetic:propionic	0	4.51	4.21	5.03	5.00	.20
	34	4.36	4.81	4.37	4.42	.20

[a]Quadratic effect of NxS fertilization (P<.05).

TABLE 22. BLOOD UREA NITROGEN CONTENT AND SERUM MINERALS OF SHEEP FED ORCHARDGRASS FERTILIZED WITH DIFFERENT LEVELS OF NITROGEN AND AN ORCHARDGRASS-RED CLOVER MIXTURE AS AFFECTED BY SULPHUR FERTILIZATION

Component	Sulphur fertilization, kg/ha	Treatments				
		N fertilization, kg/ha			Red clover	SEM
		0	224	448		
		-----------mg/dl-----------				
Blood urea nitrogen	0	24.55	26.69	29.15	20.99	1.99
	34	25.43	26.52	33.29	27.71	1.99
Serum calcium	0	1.03	1.07	1.07	1.07	.03
	34	1.08	1.10	1.06	1.09	.03
Serum magnesium	0	2.09	2.34	2.17	2.26	.07
	34	2.15	2.20	2.22	2.03	.07

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YIELD AND NUTRITIONAL VALUE OF ORCHARDGRASS AS INFLUENCED BY NITROGEN AND SULPHUR FERTILIZATION AND ASSOCIATED RED CLOVER

by

Sujatha Panditharatne

(ABSTRACT)

The effects of red clover (Trifolium pratense, L.), three levels of N (0, 224 and 448 $kg \bullet ha^{-1} \bullet yr^{-1}$, as a split application) and two levels of S (0 and 34 $kg \bullet ha^{-1} \bullet yr$) on the chemical composition and dry matter digestibility of an orchardgrass (Dactylis glomerata, L.) sward were studied. The treatments were replicated three times in a randomized block design with a split plot arrangement of treatments. Crude protein content, in vitro dry matter digestibility and dry matter yield were increased ($P<.01$) with increasing levels of N. Increasing levels of N decreased ($P<.05$) S content of orchardgrass. The inclusion of red clover did not have any significant effect on the components analyzed, probably due to a low proportion of red clover (approximately 8%) in the mixed sward. Application of S increased ($P<.05$) the S content of orchardgrass and decreased N-to-S ratio at all levels of N application. Application of S increased ($P<.05$) the crude protein and in vitro dry matter digestibility, whereas neutral detergent

fiber, lignin and hemicellulose were decreased (P<.05). Nitrogen fertilization had a quadratic effect (P<.05) on in vivo digestibility of dry matter, acid detergent fiber and hemicellulose and a linear effect (P<.001) on apparent digestibility of crude protein. Inclusion of red clover increased (P<.01) in vivo digestibility of dry matter and hemicellulose, and decreased (P<.01) digestibility of acid detergent fiber and lignin. Application of S had no direct effect on in vivo digestibility of orchardgrass. A linear interaction (P<.01) between N and S was observed for dry matter and crude protein digestibility. Rumen fluid ammonia-N was increased (P<.05) and butyric acid concentration was decreased (P<.05) linearly by N fertilization.