Carbohydrate reserves of green needlegrass (Stipa viridula Trin.) as affected by clipping and fertilization
by Larry Melvin White

A thesis submitted to the Graduate Faculty in partial fulfillment of the requirements for the degree of
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Montana State University
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Abstract:
Nitrogen (N) fertilization and frequent clipping have sometimes reduced grass stands by reducing the
carbohydrate reserves. We applied N and imposed clipping treatments to determine their effect on the
seasonal variation of total nonstructural carbohydrates (TNC) in stem bases of dryland green
needlegrass (Stipa viridula Trin.) during 1969. Ammonium nitrate was broadcast at 0, 70, and 140 kg
of N/ha in November 1968. Green needlegrass was either left unclipped in 1969 or clipped to a 5-cm
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formation (April 28). The TNC increased after second leaf formation to 19% on May 20 just before
boot stage, decreased slightly at heading (May 29) then increased to 18% at first anthesis (June 9).
Following first anthesis, TNC decreased to 9% on July 22 just prior to completion of seed
dissemination, increased to 19% in late September, decreased to 16% by November. The percentage
of TNC in clipped plants was 3 to 5 units lower at all sampling dates and the terminal value in November
was 13%. Clipping in 1969 reduced yields significantly in 1970 and tended to reduce yields in 1971.
Nitrogen increased TNC content of stem bases during mid-July and again in late autumn, apparently as
a result of new tiller development and increased photosynthesis. Nitrogen decreased the TNC only
from growth initiation until the second leaf formation. Application of N in November 1968 increased
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AS AFFECTED BY CLIPPING AND FERTILIZATION

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Approved:

K. C. Lethmer
Head, Major Department

Cochairman, Examining Committee

Cochairman, Examining Committee

Henry L. Parsons
Graduate Dean

MONTANA STATE UNIVERSITY
Bozeman, Montana

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ABSTRACT

Nitrogen (N) fertilization and frequent clipping have sometimes reduced grass stands by reducing the carbohydrate reserves. We applied N and imposed clipping treatments to determine their effect on the seasonal variation of total nonstructural carbohydrates (TNC) in stem bases of dryland green needlegrass (*Stipa viridula* Trin.) during 1969. Ammonium nitrate was broadcast at 0, 70, and 140 kg of N/ha in November 1968. Green needlegrass was either left unclipped in 1969 or clipped to a 5-cm height five times — May 7 and 28, June 17, July 8 and 29 — at approximately 21-day intervals.

A preliminary study showed that live stem bases of green needlegrass contained twice as much TNC as live roots. The coefficient of variation of TNC in live stem bases from individual plants was 20% compared to 37% for live roots.

Seasonal variation of TNC in stem bases of clipped and unclipped plants were similar, but clipped plants had a lower TNC content at all sampling dates. The TNC in non-fertilized, unclipped plants decreased from 18 to 14% from growth initiation after winter dormancy (April 5) until after second leaf formation (April 28). The TNC increased after second leaf formation to 19% on May 20 just before boot stage, decreased slightly at heading (May 29) then increased to 18% at first anthesis (June 9). Following first anthesis, TNC decreased to 9% on July 22 just prior to completion of seed dissemination, increased to 19% in late September, decreased to 16% by November. The percentage of TNC in clipped plants was 3 to 5 units lower at all sampling dates and the terminal value in November was 13%. Clipping in 1969 reduced yields significantly in 1970 and tended to reduce yields in 1971.

Nitrogen increased TNC content of stem bases during mid-July and again in late autumn, apparently as a result of new tiller development and increased photosynthesis. Nitrogen decreased the TNC only from growth initiation until the second leaf formation. Application of N in November 1968 increased yields in 1969, 1970, and 1971.
INTRODUCTION

Green needlegrass (*Stipa viridula*) is a palatable, productive range grass (Dubbs, 1966) and constitutes an important component of climax vegetation on many sites in the northern Great Plains. Improper grazing has reduced the population of this species on millions of hectares and has concomitantly reduced forage production on these ranges. Information about the effects of nitrogen (N) fertilization and clipping on carbohydrate reserves of green needlegrass is needed to improve its management.

Adequate carbohydrate reserves are important in perennial plants for winter survival, initiation of early spring growth, and initiation of regrowth after herbage removal until photosynthesis is adequate to meet plant needs. Low carbohydrate reserves limit plant growth during the first 2 to 7 days following herbage removal and this slower recovery lowers production during subsequent exponential growth (Davidson and Milthorpe, 1966b). Furthermore, tillers will die if carbohydrate reserves are decreased below the critical level (Alberda, 1966).

Although N fertilization of grasslands in the northern Great Plains generally increases forage production, it sometimes decreases production. Applications of 50 to 200 kg of N/ha on grasslands in the northern Great Plains during some drought years or frequent herbage removal reduced stands of native grasses (Klipple and Retzer, 1959), crested
wheatgrass (*Agropyron desertorum*) (Rogler and Lorenz, 1969; Seamands and Lang, 1960), and intermediate wheatgrass (*Agropyron intermedium*) (Lawrence, 1963). These stand reductions were attributed to exhaustion of carbohydrate reserves. High rates of N (200 to 400 kg of N/ha) on grasslands in the eastern United States in conjunction with frequent clipping, low soil water, and high temperature reduced stands and carbohydrate reserves of orchardgrass (*Dactylis glomerata*) (Drake et al., 1963; Celby et al., 1965) and tall fescue (*Festuca arundinacea*) (Hallock et al., 1965).

This study was initiated during the autumn of 1968 to determine the effects of N fertilization and clipping of green needlegrass. This thesis reports the effects of these treatments on the seasonal variation of carbohydrate reserves. White et al. (1972) reported the effects of the treatments on the development, growth, dry matter yield, and forage quality of green needlegrass and White and Brown (1972) reported the effects on the seasonal variation of evapotranspiration, water-use efficiency, and fertilizer N recovery.
LITERATURE REVIEW

Importance of Carbohydrate Reserves

Carbohydrate reserves are thought to be used by plants as substrate for growth and respiration. Adequate carbohydrate reserves are important in perennial plants for winter survival, early spring growth initiation, and regrowth initiation after herbage removal, when the photosynthetic production is inadequate to meet these demands. Many pasture and range management practices are based upon knowledge of how various environmental factors and herbage removal treatments affect carbohydrate reserves. This understanding helps managers to maintain high yields of desirable species and to control undesirable species.

May and Davidson (1958) and May (1960) questioned the importance of carbohydrate reserves in controlling herbage regrowth rate because only indirect evidence supported the role of reserves. However, research has shown recently that, under certain conditions, the herbage regrowth rate depends on the level of carbohydrate reserves. The following is a review of the functions of carbohydrate reserves in grasses with emphasis on recent findings. Earlier findings were summarized in the following reviews: Graber et al. (1927), Graber (1931), Weinmann (1948, 1961), Troughton (1957), May (1960), Priestley (1962), Jameson (1963), Cook (1966), and McIlroy (1967).
Reserve Constituents

Graber et al. (1927) first defined reserve energy constituents as "... those carbohydrates and nitrogen compounds elaborated, stored, and utilized by the plant itself as food for maintenance and for the development of future top and root growth." These carbohydrates, termed total available carbohydrates, are those available as energy to the plant (Weinmann, 1947). Smith (1969) suggested that the term total nonstructural carbohydrates (TNC) be used, because it is more applicable to both animal and plant investigations.

Nonstructural carbohydrates—reducing sugars (glucose and fructose), nonreducing sugar (sucrose), fructosans, and starches—are the major reserve constituents. Structural carbohydrates—hemicellulose (pentosans and hexosans) and cellulose—are not considered to provide significant reserves (McCarty, 1938; Sullivan and Sprague, 1943; Weinmann, 1948). Type, distribution in the plant and relative proportions of individual carbohydrate reserve components vary among and within grass species and under various climatic conditions during the growth season. Predominant carbohydrate reserves stored by temperate-origin grasses are sucrose and fructosans, whereas those of subtropical- or tropical-origin grasses are sucrose and starch (Cugnac, 1931; Weinmann and Reinhold, 1946; Smith, 1968; and Ojima and Isawa, 1968). The Hordeae, Aveneae, and Festuceae grass tribes store fructosan as short- or long-chain units. Genera of the Hordeae and
Aveneae tribes store predominantly short- and long-chained fructosans, respectively, while some genera of Festucae tribe characteristically store long-chained fructosans and others store short-chained fructosans (Smith, 1968).

Although Graber et al. (1927) originally defined reserve constituents as including nitrogenous compounds, most investigators have only considered carbohydrates. Recent studies indicate that proteins may be involved. Davidson and Milthorpe (1966b) concluded that nonstructural carbohydrates formed only a part of the labile pool which provided substrates for respiration and new growth of orchardgrass in a growth chamber during the first 2 to 4 days following severe herbage removal. They suggested that other labile substance, presumably proteins, must have remobilized because the amount of nonstructural carbohydrates was inadequate to account for the respiration and new growth of roots and shoots. Bilz (1966) in studying perennial ryegrass (Lolium perenne) concluded that proteinaceous material should be regarded as reserve constituents.

Most investigators generally have found that proteins are used in respiration but there is not a net utilization (Hackett, 1959). Proteinaceous sources accounted for 27 percent of the CO₂ released by respiration in phloem explants from the storage root of a carrot (Daucus carota var. sativa) (Steward et al., 1958; Bidwell et al., 1964). Breakdown products from protein turnover contributed to the
storage pool of amino acids and supplied carbon products for direct used in respiration, but carbohydrates were used preferentially over stored amino acids in synthesizing new proteins.

Studies show that even though nitrogenous compounds are used in respiration they still are not as important as carbohydrate reserves in supporting regrowth. Smith and Silva (1969) found that proportionally less nitrogenous compounds than TNC (1:18) were translocated from the roots of alfalfa (Medicago sativa) for production of new top growth after cutting in greenhouse trials. Alberda (1966) pretreated perennial ryegrass for a short period to change the plant's level of reserves. Plants with low TNC were obtained by placing them in a nutrient solution in the dark at 30 C, and plants with high TNC were obtained by placing them in water at 15 C in continuous light. The pretreatment changed the amount of nonstructural carbohydrates, but did not change the amount of organic nitrogenous compounds.

In summary, reserve constituents are those nonstructural carbohydrates which include reducing sugars, nonreducing sugars, fructosan, and starch. The predominant reserve constituents of temperate-origin grasses are sucrose and fructosan; of tropical-origin grasses, sucrose and starch. Nitrogenous compounds are used in respiration, but are not alternately stored and utilized as are carbohydrate reserves.
Nonstructural carbohydrates may be stored temporarily in all plant parts. Many scientists in the past concluded that underground organs were the major storage region for carbohydrate reserves (Weinmann, 1948; Troughton, 1957). Many other studies, however, have shown that the major storage region is generally in the stem bases (which includes stolons, corms, and rhizomes), not in the roots per se (Sampsen and McCarty, 1930; Smelov and Morazov, 1939; Sullivan and Sprague, 1943; Baker and Garwood, 1961).

The decrease of carbohydrate reserves in the roots of orchardgrass, grown in growth chambers, after severe herbage removal only accounted for less than one-tenth of root respiration (Davidson and Milthorpe, 1966b). They concluded that transfer of carbohydrate reserves from the shoots, remobilization of other substances in the roots, or both, must have occurred to account for root respiration. Marshall and Sagar (1965), using autoradiographs and labeled CO₂, found that nonstructural carbohydrates in the roots of Italian ryegrass (Lolium multiflorum) were not mobilized to the shoots to support regrowth following herbage removal, nor were labeled compounds translocated to the roots from the shoots when a part of the herbage was removed from all tillers. They concluded that "The classical view of a transference of compounds from the root to shoot following
defoliation (Troughton, 1957) ... seems unlikely ... in perennial grasses without special storage organs."

In summary, the major storage areas of carbohydrate reserves are usually the lower regions of the stems—stem bases, stolons, corms, and rhizomes. These reserves are used as an energy source to initiate new growth until photosynthesis is sufficient to sustain plant respiration. Nonstructural carbohydrates in the roots of grasses are probably not used directly in herbage regrowth following herbage removal. However, more research using labeled carbon is needed to determine if nonstructural carbohydrates in the roots are translocated aboveground for respiration or as structural components of regrowth following herbage removal.

Variation of Carbohydrate Reserves

Diurnal and seasonal. The accumulation of carbohydrate reserves in plant tissue is a dynamic system of energy balance. The level of carbohydrate reserves (hexoses and sucrose) of four grasses at Ayr of Scotland showed marked diurnal variation (Waite and Boyd, 1955). In Indiana, bromegrass (Bromus inermis) utilized almost one-third of the TNC in the herbage during the night, but diurnal fluctuations for other grass species were less (Holt and Hilst, 1969). For the grass species studied, TNC concentration in the herbage was lowest at 6 AM and increased linearly to a high at 6 PM.
The seasonal variation of carbohydrate reserves differs among grass species. In many grass species, the reserve level is lowest when the second or third leaf emerges (about one month after the start of plant growth), but in other species, the reserve level is lowest after seed ripening (Jameson, 1963). Carbohydrate reserves of Colorado wildrye (Elymus ambiguus) and mountain muhly (Muhlenbergia montana) in Colorado decreased during fast growth and increased during slow growth (McCarty, 1935). However, temperature and the availability of water and nutrients also affect the seasonal variation of carbohydrate reserves.

The accumulation of carbohydrate reserves in plant tissue is dependent upon the balance between photosynthesis and respiration. The carbohydrate reserves of orchardgrass and bermudagrass (Cynodon dactylon) grown in growth chambers decreased when growth and respiration demands were greater than photosynthetic rate and increased when growth and respiration demands were less than photosynthetic rate (Blaser et al., 1966). The level of reserves is determined by growth rate, plant development stage (Hyder and Sneva, 1959) and environment (Troughton, 1957).

Temperature. The effect of temperature on the percentage of carbohydrate reserves in the stem bases is influenced by the origin of grass species. Optimum temperatures for growth and net photosynthesis by temperate-origin grasses are about 20 to 25°C, whereas those for tropical-origin grasses are about 30 to 35°C (Evans et al., 1964; Treharne and
This difference in temperature optima for growth of two temperate species (oat \textit{(Avena sativa)} and perennial ryegrass) and two tropical species (maize \textit{(Zea mays)} and buffelgrass \textit{(Cenchrus ciliaris)}) resulted from differences in temperature optima of the major \textit{CO}_2-fixing enzymes (Treharne and Cooper, 1969). The activity of ribulose-1,5-diphosphate carboxylase is higher in temperate-origin grasses while the activity of phosphoenolpyruvate carboxylase is higher in tropical-origin grasses. Temperate-origin grasses contain only the Calvin \textit{(C}_3\textit{)} photosynthetic pathway, while tropical-origin grasses contain both the \textit{C}_4\textit{)} (Hatch and Slack) and \textit{C}_3\textit{)} photosynthetic pathways. In tropical-origin grasses, the \textit{C}_4\textit{)} pathway is located in chloroplasts of mesophyll tissue, whereas the \textit{C}_3\textit{)} pathway is located in chloroplasts of bundle sheath tissue (Berry et al., 1970; Kortschak and Nickell, 1970).

Temperature markedly affects the seasonal variation of carbohydrate reserves. Seasonal variation of total fructose in stem bases of orchardgrass (Fig. 1) was different when grown in Massachusetts, USA, than in Hokkaido, Japan (Colby et al., 1966). The total fructose level of orchardgrass grown in Hokkaido increased following heading, whereas in Massachusetts, it decreased. High June temperatures in Massachusetts apparently caused the decrease following heading. Smith and Jewiss (1966) showed that high day and night temperatures decreased the percentage of water-soluble carbohydrates in the stem bases of timothy throughout a growing season. Smith (1970) showed that changing
Fig. 1. Total fructose in the stem bases of orchardgrass plants grown in Massachusetts, USA, and Hokkaido, Japan (Colby et al., 1966).
timothy plants at inflorescence emergence from a cool to a warm regime decreased water-soluble carbohydrate content in the stem bases at early anthesis.

The effect of high day temperatures is different from that of high night temperatures. High night temperatures in a growth chamber decreased reserves of temperate-origin grasses, such as timothy (*Phleum pratense*), bromegrass, orchardgrass, and Kentucky bluegrass (*Poa pratensis*), more than high day temperatures (Baker and Jung, 1968). Increasing the day temperature, if below optimum, increases both respiration and net photosynthesis; whereas increasing the night temperature increases only the respiration rate and, thereby, decreases the reserve level. The level of carbohydrate reserves during the season may be more characteristic of climatic factors than of individual species.

Water. Eaton and Ergle (1948) in a review article noted that the effect of water stress on carbohydrate reserves varies. Some scientists have reported that drought increased the carbohydrate reserves in several grass species (Julander, 1945; Brown and Blaser, 1965; Blaser et al., 1966); others have reported that drought decreased carbohydrate reserves (Bukey and Weaver, 1939; Brown, 1943).

The degree of water stress and the plant growth stage during which it occurs will variably affect carbohydrate reserve levels. Orchardgrass under increasing water stress in a growth chamber showed a
decrease in both net photosynthetic and respiration rates (Murata and Iyama, 1963). The photosynthetic rate, however, decreased much more rapidly than respiration, thus lowering carbohydrate reserves. If water stress stops stem elongation and has only minor effects on photosynthesis, as reported by Wardlaw (1968), carbohydrate reserves would then increase. Brown and Blaser (1970) suggested that the build-up of carbohydrate reserves and inorganic nitrogen in plants under water stress results from the transformation of carbon-containing nitrogenous substances.

Nitrogen. The effects of N fertilization on carbohydrate reserves are complex and variable. Weinmann (1948) in a review article noted cases where N fertilization caused no effect, increased, or decreased carbohydrate reserves. Generally, N applied at low to moderate rates increases carbohydrate reserves. Nitrogen applied at high rates decreases carbohydrate reserves (Adegbola and McKell, 1966). The physiological reasons why N variably affects carbohydrate reserves are not well understood.

If N is deficient, application of moderate amounts of N can increase plant growth when carbohydrates, water, and other nutrients are available and environmental conditions are favorable. Increased plant growth from N application was associated with increased leaf area, chloroplast protein, and chlorophyll content which increased photosynthesis (Murata, 1969). The increased photosynthetic activity can
then, theoretically, increase carbohydrate reserves.

Excess N tends to decrease carbohydrate reserves when other nutrients and environment do not limit plant growth. In this case, N fertilization stimulates the synthesis of amino acids and amide compounds to the detriment of carbohydrate reserves (Prianishnikov, 1951). Carbohydrate reserves are used as the carbon-skeleton for protein synthesis (Prianishnikov, 1951).

Application of high rates of N fertilizer (200 to 400 kg N/ha) in conjunction with frequent clipping, low soil water, and high temperatures reduced stands and carbohydrate reserves of orchardgrass in Massachusetts (Drake et al., 1963; Colby et al., 1965) and tall fescue in Virginia (Hallock et al., 1965). Applications of high rates of N also reduced stands of orchardgrass and tall fescue in Maryland (Alexander and McCloud, 1962).

Scientists also reported that application of 50 to 200 kg N/ha reduced severely the grass stand, especially when associated with drought or frequent herbage removal on native range in Colorado (Klipple and Retzer, 1959) on crested wheatgrass in Wyoming (Seamands and Lang, 1960) and North Dakota (Rogler and Lorenz, 1969), and on intermediate wheatgrass in Saskatchewan (Lawrence, 1963). These scientists assumed that carbohydrate reserves were reduced. Frequent clipping, with or without N fertilization, decreased the percentage basal ground cover of non-irrigated green needlegrass in Saskatchewan.
(Heinrichs and Clark, 1961). In contrast, N fertilization (0 to 375 kg N/ha) did not increase the winter-kill of intermediate wheatgrass under irrigation in Saskatchewan, although frequent and close clipping did (Lawrence and Ashford, 1969).

High rates of N should not be applied under the combined conditions of drought and high temperatures. Under these conditions, clipping or grazing could deplete carbohydrate reserves below a critical level, and cause stand reduction and poor growth recovery.

In summary, the interaction of the plant with the environment and the balance between photosynthesis and respiration determine the variation of carbohydrate reserves during the growing season. In some grass species, a low reserve occurs when the second or third leaf emerges; in other grasses, it may occur just before or after seed ripening. The seasonal variation of carbohydrate reserves can differ for the same species grown in different environments. Above-optimum temperatures, especially during the night, decrease carbohydrate reserves; whereas water stress can either increase or decrease reserves, depending on the degree of stress and stage of plant growth.

Studies to date generally show that N applied at low to moderate rates increases carbohydrate reserves, but that N at moderate to high rates decreases reserves. Excess N applied during periods of water stress and high temperatures, coupled with frequent herbage removal, often reduces stands and growth rate. More research is needed to
fully understand the effects of N fertilization on carbohydrate reserves.

**Regrowth After Partial Herbage Removal**

**Clipping.** The effect of herbage removal on plant regrowth has been classified into three simplified categories. Herbage removal reduces (1) amount of carbohydrate reserves, (2) root growth, and (3) leaf area (Alcock, 1964). Many other factors, however, also affect the regrowth rate of a sward following herbage removal (Jameson, 1963).

The importance of carbohydrate reserves in controlling regrowth rate following herbage removal is a controversial topic in grassland management. The results of many field studies show that carbohydrate reserves decreased in both stem bases and roots of grasses after cutting (Graber et al., 1927; Troughton, 1957) until sufficient leaf area developed that carbohydrates produced in photosynthesis equaled those used in respiration and growth. This observation led to the general belief that, following herbage removal, some reserves are converted to structural components of the new and expanding cells, and thus the reserve level affects the regrowth rate. However, May (1960) stated that the "... specific role for carbohydrate reserves in initiating regrowth, and in determining the rate or ultimate extent of regrowth, cannot yet be considered as firmly established."

Since May's (1960) conclusion, new research techniques have
provided evidence that carbohydrate reserves are used for regrowth following herbage removal. Carbohydrate reserves, assimilated as labeled CO$_2$ by bahiagrass (Paspalum notatum) were used to help form leaves for 6 days after herbage removal (Ehara et al., 1966). Those nonstructural carbohydrates in alfalfa (labeled by $^{14}$CO$_2$ assimilation) which were initially stored in the root and crown were utilized after herbage removal as substrate for respiration of both roots and tops and as structural components for top growth (Pearce et al., 1969; Smith and Marten, 1970).

The dry weight of perennial ryegrass with high carbohydrate reserves did not increase for 4 days following clipping because respirational losses exceeded photosynthetic gain but the weight of plants with low carbohydrate reserves did not increase for 7 days (Alberda, 1966). Davidson and Milthorpe (1965, 1966a, 1966b) after measuring both respiration and photosynthetic rates of orchardgrass grown in growth chambers, concluded that regrowth following clipping depended upon carbohydrate reserves for only the first 2 to 4 days. During this period, stored carbohydrates were used for regrowth and respiration. Afterwards, regrowth depended on other factors, such as photosynthetic rate and nutrient uptake. Milthorpe and Davidson (1966) demonstrated that, even though carbohydrate reserves influenced the regrowth rate for only the first few days, the initial stimulus due to the level of reserves was maintained during subsequent exponential growth. Measure-
ment of the initial response attributable to the level of carbohydrate reserves during the exponential phase of orchardgrass regrowth is difficult because complex interrelationships frequently obscure the response (Davidson and Milthorpe, 1966b).

Ward and Blaser (1961) concluded that carbohydrate reserves of orchardgrass in Virginia stimulated dry matter production for the first 25 days after partial or complete herbage removal; thereafter, regrowth rates were dependent on leaf area. Davidson and Milthorpe (1966b) re-examined Ward and Blaser's data and concluded that the relative rate of leaf expansion of plants with high and low levels of carbohydrate reserves was the same and the effect of carbohydrate reserve levels was confined only to the initial stage of regrowth.

Weinmann (1948) stated that clipping does not always reduce carbohydrate reserves, and Jameson (1963) stated that regrowth rate does not always depend upon the level of carbohydrate reserves. Such discrepancies may be due to any of the following factors: (1) Variation in amount or capacity of photosynthetic tissue remaining after herbage removal; (2) sampling for reserves too late after clipping, when the reserves have already been restored; and (3) sampling the wrong plant part.

Photosynthetic capacity remaining after herbage removal depends on height of cutting, growth habit of the plant, and age of remaining leaves. The photosynthetic rate of the sheath of young orchardgrass
tillers grown in growth chambers was about one-third of the rate of the blades (Davidson and Milthorpe, 1966b). The maximum photosynthetic rate of a leaf occurred when the leaf blade first emerged from the sheath and this rate then decreased with age in tall fescue (Jewiss and Woledge, 1967) and orchardgrass (Treharne et al., 1968). The life span of an orchardgrass leaf in Kentucky, after it reached full extension, was only about 28 days (Taylor and Templeton, 1966). Thus leaf blades about 28 days after full extension would be of little value in maintaining the photosynthetic rate of a sward.

In summary, the level of carbohydrate reserves in the lower regions of the stems apparently affects the regrowth rate for 2 to 7 days after herbage removal; but this initial support from carbohydrate reserves can be maintained during subsequent exponential growth. After the initial period, plant regrowth rate depends on other factors, such as leaf area and nutrient uptake.

Grazing versus clipping. The effects of grazing and clipping are similar, but not identical. Clipping removes all herbage above a given height from all plants, whereas grazing removes herbage at heights varying from plant to plant and even within the same plant. Hormay and Talbot (1961) reported that grazing of Idaho fescue (*Festuca idahoensis*) by cows in an opening of ponderosa pine (*Pinus ponderosa*) in California was not uniform from plant to plant. When the overall utilization of Idaho fescue herbage was 43%, 40% of the plants were grazed to a 2.5-cm
stubble height; 29%, to 5.1-cm; 13%, to 7.6-cm; and 3%, to 10.2-cm or taller; and 15% were not grazed at all.

Grazing reduces plant vigor more than clipping at the same degree of herbage removal because grazing often removes herbage from one plant and not from the surrounding vegetation. In Montana, clipping of individual Idaho fescue plants and not the surrounding vegetation reduced yields of clipped plants more the following year than when the surrounding vegetation was also clipped (Mueggler, 1970). Unclipped plants competed advantageously with clipped plants for nutrients and water.

Grazing, however, can be less detrimental to plant vigor than clipping by leaving ungrazed tillers in a plant. Carbohydrate reserves of tall fescue in Missouri (as indicated by greater etiolated regrowth) increased as the percentage of unclipped tillers per plant increased from 0 to 30% (Matches, 1966). Carbohydrate reserves of dallisgrass (Paspalum dilatatum) in Mississippi also increased as the percentage of unclipped tillers increased from 0 to 10% (Watson and Ward, 1970).

Carbohydrate compounds are transferred from unclipped to clipped tillers only for the first few days following cutting and only in small plants. When a single unclipped tiller remained, it initially transported labeled carbon products to defoliated tillers within the same plant (Marshall and Sagar, 1965). However, transfer of assimilates from an unclipped tiller to clipped tillers did not occur beyond the third
day after cutting. More mature tillers of Italian ryegrass were independent because leaf blades fed labeled CO\textsubscript{2} transported labeled assimilates only to the root system of that tiller (Marshall and Sagar, 1965). Tillers of large plants of weeping lovegrass (\textit{Eragrostis curvula}) in Maryland apparently were not completely connected together by vascular connections at the crown because partial clipping of a group of tillers stopped root growth only of clipped tillers (Crider, 1955). Leaving unclipped tillers not only increased production because of the transfer of carbohydrates but also because of leaving photosynthetic tissue and carbohydrate reserves stored in the stems.

In summary, the effect of grazing on herbage regrowth rate can be more or less detrimental than clipping, depending on circumstances. Grazing may be more detrimental than clipping if grazing removes all herbage from some plants and not others, because ungrazed plants take available nutrients and water away from grazed plants. However, grazing may be less detrimental than clipping if grazing leaves ungrazed tillers on a plant while removing others, thus allowing for the transfer of carbohydrates from ungrazed to grazed tillers.

**Management Implications**

Various management practices—range readiness, season of use, degree of utilization, and grazing systems—are partially based upon how they affect carbohydrate reserves of grasses (National Research
Effects of a particular management practice often can be evaluated in a single year by observing carbohydrate reserve levels and variation. The effects of various management practices on plant vigor can be partially measured objectively and quantitatively with the percentage of TNC (Cook, 1966; National Research Council, 1962).

The seasonal variation of carbohydrate reserves of many grasses has not been determined. Knowledge of the seasonal variation of carbohydrate reserves and the effects of climate and management practices on them will help pasture and range managers improve present management practices. Factors other than carbohydrate reserves—leaf area, light interception, root area, nutrient uptake, competition and other morphological and physiological factors—also influence the effects of herbage removal.

Cook (1966) stated that "Proper management . . . does not necessarily imply that a maximum level of carbohydrate reserves be maintained, but care must be taken . . . that these reserves do not fall below a critical level" or tillers will die. More research is needed to determine critical levels of carbohydrate reserves at which some tillers die. Perennial ryegrass grown in growth chambers was unable to use carbohydrate reserves below 6% of dry weight, the amount required for normal cell function; and at the 6% level, reserves inadequately supported the existing tiller population and some tillers
died (Alberda, 1966; del Pozo, 1963). The critical level of carbohydrate reserves can be different among grass varieties (Davies, 1965) and species and is probably affected by fertility, management, environment, and season.
STUDY AREA

This study was conducted from 1968 through 1971 on a solid stand of green needlegrass seeded in 1961. The site is 11 km north of Culbertson, Mont. on the Soil and Water Conservation District Research Farm. The soil is a Williams loam (sandy loam) derived from glacial till, sloping from 4 to 6% to the south. In July 1968, 44.8 kg of phosphorus/ha was broadcast uniformly, since the soil is normally deficient in phosphorus. After application of phosphorus, the soil contained over 5 ppm of available phosphorus to a depth of 30 cm (White, 1971). The results of chemical analysis of this soil are shown in Table 1. The study area is classified as a sandy 25.4- to 35.6-cm precipitation zone range site. Prior to 1969, green needlegrass herbage was harvested annually for hay when the first seeds ripened.

Weeds in the stand, primarily volunteer alfalfa, were controlled by spraying with 1.1 kg/ha of active ingredient 2,4-D amine on May 12, 1969. From 1969 through 1971, leaf spot disease, Alternaria tenuis, infected the entire second leaf and the distal half of the third leaf during the middle of May causing light to moderate damage. Subsequent infections did not occur during the growing season of any year.

The experimental site normally receives 32.6 cm of precipitation annually, 26.0 cm of precipitation during the growing season (April
Table 1. Summary of soil analysis data (July 1968) before applications of N-P fertilizer.

<table>
<thead>
<tr>
<th>Depth</th>
<th>pH</th>
<th>Soil texture</th>
<th>Conductivity</th>
<th>Organic Available P (Bray-HCL)</th>
<th>Available K</th>
<th>Available nitrate*</th>
</tr>
</thead>
<tbody>
<tr>
<td>0 - 7.5</td>
<td>7.4</td>
<td>s1</td>
<td>0.8</td>
<td>2.1</td>
<td>23</td>
<td>390</td>
</tr>
<tr>
<td>7.5 - 15</td>
<td>7.1</td>
<td>s1</td>
<td>0.7</td>
<td>1.5</td>
<td>6</td>
<td>250</td>
</tr>
<tr>
<td>15 - 30</td>
<td>7.2</td>
<td>s1</td>
<td>0.7</td>
<td>1.2</td>
<td>6</td>
<td>160</td>
</tr>
<tr>
<td>30 - 60</td>
<td>7.7</td>
<td>s1-cl†</td>
<td>1.1</td>
<td>1.1</td>
<td>4</td>
<td>130</td>
</tr>
</tbody>
</table>

*Average of values measured in May and August 1969 (nonfertilized plots).
†Sandy loam-clay loam.
to September), 114 frost-free days, and the mean annual air temperature is 5.2 °C. During the 1969 growing season, above-normal precipitation supplied adequate soil water from August 1968 through April 1969, and from June 25 through early August 1969. However, below-normal precipitation and above-normal temperatures caused drought stress which retarded the growth rate in plants from the last of May through June 24 and again from mid-August through mid-September 1969 (White, and Brown, 1972). Precipitation during the 1970 growing season was above normal except for the last three weeks of May and first two weeks of June. Precipitation during the 1971 growing season was below normal and herbage production was only about one-third of normal.
METHODS

Experimental Design and Treatments

Factorial combinations of three levels of N and two levels of clipping were assigned to plots in a completely randomized split-plot design with four replications. Ammonium nitrate was broadcast on main plots (17 by 5 m) in November 1968 at 0, 70, and 140 kg of N/ha. The two clipping treatments applied on subplots (8.5 by 5 m) were (A) plants undisturbed until sampled (unclipped) and (B) plants clipped to a 5-cm height at about 21-day intervals on May 7 and 28, June 17, July 8 and 29, 1969.

Total Nonstructural Carbohydrates

A preliminary study was conducted during October 1968, to determine the plant part and number of samples required to adequately determine the percentage of TNC in green needlegrass. Samples included stem bases to a 5-cm height aboveground and roots in a 15-cm diameter by 15-cm deep core. Individual cores were washed free of soil and the plant material was separated as stem bases or roots and as either live or dead. Material from 35 individual plants was analyzed for TNC to determine the variability among plants. Also, ten plants from each subplot were composited and analyzed for TNC to determine the variability of composited samples.
Only live stem bases were sampled for TNC in 1969. A sample of 10 plants from each subplot was taken for analysis at about 10-day intervals from April 8 through August 12, on September 2 and 27, and on November 8, 1969. From April 8 through May 8 (four sampling dates), only the plants on unclipped subplots were sampled, since clipping did not begin until May 7. Herbage above 5 cm was clipped and discarded. Blasts of air were used to remove soil from the stem bases. Within one-half hour after sampling, samples were placed in a forced-air oven at 90°C for one hour to stop enzymatic action and were then dried at 70°C to constant weight. Roots and all dead leaf sheaths (dead at the time of sampling) were removed. Dry stem bases were weighed to the nearest tenth of a gram and ground to pass a 40-mesh screen, then redried at 70°C and stored in glass jars for subsequent analysis.

The stem bases were analyzed for TNC following the procedure described by Smith (1969). The TNC was extracted and hydrolysed with takadiastase enzyme solution (Clarase 900, Miles Laboratories, Inc., Elkhart, Ind.) because green needlegrass stores some starch (Smith, 1968). Reducing power was measured by the Shaeffer-Somogyi copperiodometric titration method as described by Heinze and Murneek (1940). Results were reported as a percentage of dry weight using a glucose standard.
Other Analyses

Stem bases were also analyzed for total N by the Kjeldahl procedure (Jackson, 1958). The residual effects of N fertilization in November 1968 and clipping in 1969 upon dry matter yield in 1970 and 1971 were determined from samples harvested when seed was ripe. Dry matter was harvested from two permanent ¼- by 4-m areas by clipping plants to a 5-cm height. Within treatment A, samples were collected from subplots which had not been clipped in 1969. Within treatment B, samples were collected from the same permanent plots used in 1969.

Soil water content (0- to 120-cm depth) was determined approximately every 10 days from April through August 1969 as previously described (White and Brown, 1972). Minimum and maximum daily air temperatures were measured at a standard weather station adjacent to the plots.

Statistical Analyses

Standard analysis of variance was used to determine if treatment differences were significant. When the F-test for N means was significant at the 5% probability level, Duncan's Multiple Range test was used to determine which differences among N means were significant. All differences discussed are significant at the 5% probability unless stated otherwise.

When subsequent measurements were made over a period of time, time was considered an additional variable; therefore, data were
analyzed as split in time. When date by clipping or date by N interaction was significant at the 5% level, analysis of variance was made for each date. When error (B) was larger than error (A), the two error terms were pooled before the F-tests were made (Steel and Torrie, 1960).
RESULTS

Preliminary Study

The preliminary study conducted during October 1968 showed that live stem bases contained 13.9% TNC and dead stem bases only 3.4% TNC. The TNC in material designated as dead stems was disproportionately high due to the inclusion of some current year's spring tillers which were difficult to distinguish from dead stems. Live roots contained 6.7% and dead roots only 0.9% TNC.

Analysis of individual plants for TNC showed that the coefficient of variation for live stem bases was 20% and for live roots, 37%. It was estimated that 16 individual plants were needed in a sample for live stem bases and 52 individual plants for live roots to obtain a sample mean within 10% of the population mean at the 95% probability level. When plots were arranged in a completely randomized design with four replications and 10 plants were composited per sample, the coefficient of variation was 16.5% for live stem bases (Table 2) and 21.7% for live roots.

From information obtained in the preliminary study, 10 individual plants were composited per sample from each subplot in 1969. Compositing 10 plants per subplot, when main plots were arranged in a completely randomized design with four replications, resulted in coefficients of variation for the percentage of TNC in live stem bases at individual sampling dates ranging from 7 to 16% for both N and clipping treatments.
Table 2. Analysis of variance of percent TNC in stem bases of green needlegrass during October 1968.

<table>
<thead>
<tr>
<th>Source of Variance</th>
<th>df</th>
<th>SS</th>
<th>MS</th>
<th>F Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Nitrogen</td>
<td>2</td>
<td>9.83</td>
<td>4.91</td>
<td>0.94NS</td>
</tr>
<tr>
<td>Clipping</td>
<td>1</td>
<td>14.73</td>
<td>14.73</td>
<td>2.81NS</td>
</tr>
<tr>
<td>Nitrogen x Clipping</td>
<td>2</td>
<td>9.59</td>
<td>4.79</td>
<td>0.91NS</td>
</tr>
<tr>
<td>Error</td>
<td>18</td>
<td>94.30</td>
<td>5.24</td>
<td></td>
</tr>
<tr>
<td>Total</td>
<td>23</td>
<td>128.45</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

C.V. = 16.5%
Plant Growth

Growth of the 1969 crop of green needlegrass started in late August 1968 after the heavy mid-August rains. White et al. (1972) estimated that two-thirds of the floral tillers present in July 1969 were initiated the previous autumn. Tiller initiation which started in the autumn of 1968 continued during the spring of 1969 after the spring thaw (April 5). Table 3 shows average dates when green needlegrass reached given stages of development.

The growth of green needlegrass during 1969 is illustrated by changes in the weight of stem bases per 10 plants (Fig. 2). Limited soil water from late May through mid-June (Fig. 3) probably stopped tiller initiation during this period. However, stem base weight of unclipped plants increased during early June because of elongation and enlargement of the stems of floral tillers. Rains received the last of June and early July stimulated tiller initiation and growth during July and early August. White et al. (1972) showed that the number of tillers taller than 5 cm increased during July. Tillers less than 5 cm were not counted during this period. However, limited soil water (Fig. 3) again stopped tiller initiation and growth from the middle of August through early September (Fig. 2). The 2.74 cm of rain received September 21 again stimulated growth and the weight of stem bases increased from the last of September through October.
Table 3. The date unclipped autumn-initiated (1968) floral tillers of green needlegrass reached successive developmental stages during 1969 (White et al., 1972).

<table>
<thead>
<tr>
<th>Developmental Stages</th>
<th>Date</th>
</tr>
</thead>
<tbody>
<tr>
<td>Tillers started growth</td>
<td>August 15, 1968</td>
</tr>
<tr>
<td>Emergence of first leaf*</td>
<td>September 1968</td>
</tr>
<tr>
<td>Over-wintering</td>
<td>Winter 1968-69</td>
</tr>
<tr>
<td>Start of spring growth</td>
<td>April 5, 1969</td>
</tr>
<tr>
<td>Emergence of second leaf</td>
<td>April 22, 1969</td>
</tr>
<tr>
<td>Emergence of third leaf</td>
<td>May 12, 1969</td>
</tr>
<tr>
<td>Head in-boot</td>
<td>May 26, 1969</td>
</tr>
<tr>
<td>First head appearance</td>
<td>May 29, 1969</td>
</tr>
<tr>
<td>Emergence of fourth leaf</td>
<td>June 2, 1969</td>
</tr>
<tr>
<td>First anthesis</td>
<td>June 9, 1969</td>
</tr>
<tr>
<td>Full anthesis</td>
<td>June 12, 1969</td>
</tr>
<tr>
<td>Milk stage</td>
<td>June 17, 1969</td>
</tr>
<tr>
<td>Soft dough stage</td>
<td>June 24, 1969</td>
</tr>
<tr>
<td>Hard dough stage</td>
<td>July 7, 1969</td>
</tr>
<tr>
<td>First seed ripe</td>
<td>July 7, 1969</td>
</tr>
<tr>
<td>Start of seed dissemination</td>
<td>July 7, 1969</td>
</tr>
<tr>
<td>Completion of seed dissemination</td>
<td>July 26, 1969</td>
</tr>
</tbody>
</table>

*Leaf collar appears.
Fig. 2. Weight of stem bases (averaged over N levels) from ten green needlegrass plants during 1969 when clipped five times from May through July 1969 or left unclipped.
Fig. 3. Five-day mean air temperatures and mean soil water content in the 0- to 30- and 30- to 60-cm depths during 1969.
Analysis of TNC Data

The analysis of variance of the TNC data for the first period of 1969, April 8 to May 8, was made separately from that of the second period of 1969, May 20 to November 8. During the first period, the clipping treatments had not yet been applied, therefore only plants on the unclipped plots were sampled for TNC. The N by date interaction for TNC data during the first period was highly significant (Table 4) and the N by clipping by date interaction during the second period was highly significant (Table 5). Therefore, neither the N by date means nor the clipping by date means were used to interpret TNC data. The effects of N rate and clipping frequency on TNC data were analysed using only the averages of the data over replications.

Seasonal Variation of TNC

The concentration of TNC in the stem bases of unclipped green needlegrass varied considerably during the growing season and was related to growth stage, Fig. 4. The percentage of TNC in non-fertilized, unclipped plants decreased from 18% after the start of spring growth on April 5 to 14% on April 28 after the emergence of the second leaf. The TNC accumulated rapidly after emergence of the second leaf and reached about 19% on May 20 just before the boot stage, then decreased slightly with heading (May 29) then increased to 18% at first anthesis (June 9). The TNC declined after anthesis to
Table 4. Analysis of variance of percentage of TNC in stem bases of green needlegrass from April 8 through May 8, 1969.

<table>
<thead>
<tr>
<th>Source of Variance</th>
<th>df</th>
<th>SS</th>
<th>MS</th>
<th>F Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Nitrogen</td>
<td>2</td>
<td>68.28</td>
<td>34.14</td>
<td>26.56**</td>
</tr>
<tr>
<td>Error (A)</td>
<td>9</td>
<td>11.57</td>
<td>1.28</td>
<td></td>
</tr>
<tr>
<td>Dates</td>
<td>3</td>
<td>86.06</td>
<td>28.69</td>
<td>19.41**</td>
</tr>
<tr>
<td>Nitrogen x Dates</td>
<td>6</td>
<td>29.76</td>
<td>4.96</td>
<td>3.35**</td>
</tr>
<tr>
<td>Error (B)</td>
<td>27</td>
<td>39.90</td>
<td>1.48</td>
<td></td>
</tr>
<tr>
<td>Total</td>
<td>47</td>
<td>235.57</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

C. V. (A) = 7.5%
C. V. (B) = 8.0%
Table 5. Analysis of variance of percentage of TNC in stem bases of green needlegrass from May 20 through November 8, 1969.

<table>
<thead>
<tr>
<th>Source of Variance</th>
<th>df</th>
<th>SS</th>
<th>MS</th>
<th>F Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Nitrogen</td>
<td>2</td>
<td>71.94</td>
<td>35.97</td>
<td>6.69*</td>
</tr>
<tr>
<td>Error (A)</td>
<td>9</td>
<td>48.37</td>
<td>5.37</td>
<td></td>
</tr>
<tr>
<td>Clipping</td>
<td>1</td>
<td>759.50</td>
<td>759.50</td>
<td>668.59**</td>
</tr>
<tr>
<td>Nitrogen x Clipping</td>
<td>2</td>
<td>15.65</td>
<td>7.82</td>
<td>6.88*</td>
</tr>
<tr>
<td>Error (B)</td>
<td>9</td>
<td>10.22</td>
<td>1.14</td>
<td></td>
</tr>
<tr>
<td>Dates</td>
<td>11</td>
<td>1415.58</td>
<td>128.69</td>
<td>64.41**</td>
</tr>
<tr>
<td>Nitrogen x Dates</td>
<td>22</td>
<td>231.74</td>
<td>10.53</td>
<td>5.27**</td>
</tr>
<tr>
<td>Clipping x Dates</td>
<td>11</td>
<td>131.14</td>
<td>11.92</td>
<td>5.97**</td>
</tr>
<tr>
<td>N x C x D</td>
<td>22</td>
<td>125.30</td>
<td>5.70</td>
<td>2.85**</td>
</tr>
<tr>
<td>Error (C)</td>
<td>198</td>
<td>395.59</td>
<td>2.00</td>
<td></td>
</tr>
<tr>
<td>Total</td>
<td>287</td>
<td>3205.04</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

C. V. (A) = 15.5%, C. V. (B) = 7.1%, C. V. (C) = 9.5%
Fig. 4. Percentage of TNC in stem bases of unclipped green needlegrass during 1969 with and without N fertilization during 1968.
a low of 9% on July 22 just prior to completion of seed dissemination. Following seed dissemination, TNC increased to 19% in late September and then decreased to 16% in November.

Effects of Clipping

Clipping five times in 1969 reduced the TNC level an average of 3 to 5 units lower at all sampling dates than that of unclipped plants (Fig. 5); however, the seasonal variation of TNC was similar for both treatments. The TNC of clipped plants decreased to a low of 9% on July 10, which was 12 days earlier than the lowest TNC level in unclipped plants. The TNC of clipped plants increased similarly to unclipped plants after July 22. The terminal TNC value (November) of clipped plants was 13% in contrast to 17% for unclipped plants.

The effect of clipping was hard to distinguish following the third clipping because the TNC level 13 days after clipping increased at about the same rate in both unclipped and clipped plants. However, clipping significantly reduced the TNC level on all dates except on May 29 and July 22. The lack of a significant reduction of TNC by clipping of May 29 and July 22 was associated with increased herbage production during these periods (White et al., 1972). Greater herbage production would be associated with more leaf area and in turn should increase the photosynthetic capacity of the plants.

Clipping in 1969 reduced the weight of stem bases during 1969 by an average of 30% for the season (Fig. 2). White et al. (1972).
Fig. 5. Mean percentage of TNC (averaged over N levels) in stem bases of green needlegrass during 1969 when clipped five times from May through July 1969 or left unclipped.
reported that clipping in 1969 decreased dry matter yield during 1969 by 24%. The effect of clipping appeared to persist in 1970 and 1971. Clipping significantly decreased dry matter yield by 24% in 1970 (Table 6) and also reduced the inflorescence height by 20% in 1970 (White et al., 1972). Clipping appeared to decrease dry matter yield by 12% in 1971 (not significant).

Effects of N Fertilization

Unclipped plants. When growth began April 5, the percentage of TNC in the stem bases of N-fertilized, unclipped plants was less than in stem bases of non-fertilized, unclipped plants (Fig. 4). The TNC in N-fertilized plants decreased to its lowest spring level earlier than in non-fertilized plants. From April 28 to May 20, TNC accumulated rapidly in plants on all N treatments. Nitrogen fertilization increased the percentage of TNC at inflorescence emergence (May 29) and at first anthesis (June 9). Because soil water (Fig. 3) was apparently limiting plant growth, N fertilization did not affect TNC on June 19. However, the 140 kg of N decreased TNC on June 30 compared to that of the 0 and 70 kg of N rates. Application of 140 kg of N/ha initiated tiller growth (increased stem-base weight) earlier after the late June rains than either the 0 or 70 kg of N rate (Fig. 6), and this may have been associated with the TNC decrease on June 30.

<table>
<thead>
<tr>
<th>Clipping Frequency</th>
<th>N rate, kg/ha</th>
<th>Avg.</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>0</td>
<td>70</td>
</tr>
<tr>
<td>Undipped</td>
<td>640</td>
<td>1130</td>
</tr>
<tr>
<td>Clipped</td>
<td>400</td>
<td>920</td>
</tr>
<tr>
<td>Avg.</td>
<td>520 c</td>
<td>1025 b</td>
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<td>Clipped</td>
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<td>Avg.</td>
<td>190 a</td>
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*Average values for clipping treatments or for nitrogen treatments followed by the same letter are not significantly different at the 5% level of probability.
Fig. 6. Weight of stem bases from ten unclipped green needlegrass plants during 1969 with and without N fertilization during 1968.
The magnitude of TNC decrease after anthesis was inversely proportional to N-fertilization rate and was apparently related to new tiller formation. Nitrogen fertilization increased the number of new tillers formed following rains in late June and early July. From July 2 to 29, tiller numbers increased 0, 2.5, and 37.0% on plots fertilized with 0, 70, and 140 kg of N/ha, respectively (White et al., 1972). The 140 kg of N increased the weight of stem bases earlier than did the 70 kg of N following the late June and early July rains (Fig. 6). The 70 kg of N increased the TNC on August 12 over that of the 0 kg of N while the 140 kg of N did not. The 70 kg of N may have increased the TNC because it initiated tiller growth later than did the 140 kg of N.

The TNC concentrations remained equal among N-fertilizer treatments from mid-August through mid-September when soil water tension at 0 to 30 cm exceeded the wilting point (Fig. 3). When growth resumed after a September 21 rain, TNC decreased in the stem bases of non-fertilized plants and in plants fertilized with 70 kg of N. However, the TNC in stem bases of plants fertilized with 140 kg of N did not decrease.

Clipped plants. From May 20 to July 10, TNC of non-fertilized plants declined in a linear manner from 17 to 7.5% (Fig. 7). In contrast, the decline of TNC in fertilized plants was more variable. Nitrogen fertilization did not affect TNC of clipped plants when sampled two days after clipping. However, approximately 13 days after clipping, N appeared to increase (not significant) the TNC on May 20.
Fig. 7. Percentage of TNC in stem bases of frequently clipped green needlegrass during 1969 with and without N fertilization during 1968.
and June 9, and significantly increased TNC on June 30, July 22, and August 12.

The 140 kg of N increased TNC of clipped plants on July 22 and the 70 kg of N increased TNC on August 12. The 140 kg of N probably increased TNC earlier because it increased tiller initiation (stem base weight) earlier than the 70 kg of N following the late June and early July rains (Fig. 8).

The TNC level of clipped plants on all N treatments decreased following autumn growth which began after the September 21 rain. The decrease in TNC was greater in plants not fertilized or fertilized with 70 kg of N than in plants fertilized with 140 kg of N.

Residual effects of the November 1968 application of N were present in both 1970 and 1971. In 1970, residual N from the 70 kg and 140 kg of N applications increased dry matter yield 100 and 123%, respectively (White et al., 1972). Also residual N from the 140 kg of N treatment increased the inflorescence height in 1970 by 15% more than did either the 0 or 70 kg of N (White, 1971). In 1971, the residual from the 140 kg of N application increased the dry matter yield by 130% (Table 6). Residual N from 70 kg of N appeared to increase the dry matter yield by 50% (not significant).
Fig. 8. Weight of stem bases from ten frequently clipped green needlegrass plants during 1969 with and without N fertilization during 1968.
Total Nitrogen

In the stem bases of unclipped green needlegrass, the percentage of total N (Fig. 9) varied inversely with the percentage of TNC (Fig. 4) during the spring and early summer. During the remainder of the season, total N concentrations remained near 1.5% while TNC concentrations fluctuated greatly.

Frequent clipping of green needlegrass increased the percentage of total N (Fig. 10) during July and at the same time decreased the TNC level (Fig. 5) in the stem bases. The increase in percentage of total N (Fig. 10) during June and July by clipping was accentuated by a decrease in dry weight of the stem bases (Fig. 2). Also, the increased N during July may have resulted because clipping extended the period of plant growth (White et al., 1972), and resulted in N uptake for a longer period. Clipping had no effect on the percentage of total N in the stem bases during the autumn of 1969. Thus, clipped plants at the end of the growing season of 1969 had the same percentage of total N as unclipped plants but less TNC for winter survival and spring growth initiation.
Fig 9. Percentage of N in stem bases of unclipped green needlegrass during 1969 with and without N fertilization during 1968.
Fig. 10. Mean percentage of N (averaged over N levels) in stem bases of green needlegrass during 1969 when clipped five times from May through July 1969 or left unclipped.
DISCUSSION

Seasonal Variation of TNC

The TNC concentrations always decrease with early growth until photosynthesis in exposed leaves can supply the energy required for respiration and growth. The rate, duration, and amount of this TNC decrease vary among species and environmental conditions. When green needlegrass had produced 15% of its maximum annual upstretched-leaf height, it had attained sufficient leaf area and photosynthetic capacity to stop carbohydrate reserves from decreasing further. Blue-bunch wheatgrass (Agropyron spicatum) had produced 45% (McCarty and Price, 1942), and mountain bromegrass (Bromus marginatus), only 10% of its maximum annual upstretched-height before carbohydrate reserves ceased decreasing (McIlvaine, 1942). Green needlegrass is somewhat like crested wheatgrass, since it started increasing its TNC level earlier than bluebunch wheatgrass because of earlier leaf development, smaller percentage of floral tillers, and later initiation of stem elongation (Hyder and Sneva, 1959).

The TNC decrease in late May coincided with higher air temperatures (Fig. 3) and rapid elongation of floral tillers during the booting stage. The carbohydrate reserves of perennial ryegrass, orchardgrass, meadow fescue (Festuca pratensis), and timothy also decreased during the period of rapid elongation of floral tillers
Waite and Boyd (1953). Hyder and Sneva (1959) found that removing the inflorescence from crested wheatgrass at emergence did not stop carbohydrate reserves from decreasing in a manner similar to that in plants allowed to develop seed.

The TNC level (Fig. 4) during the last of May and early June appeared to fluctuate inversely with air temperature (Fig. 3). The TNC decreased during periods with higher temperatures and increased during periods with lower temperatures. Among others, Brown (1939) working with Canada bluegrass (Poa compressa), Kentucky bluegrass, and orchardgrass, and also Sullivan and Sprague (1949) working with perennial ryegrass, found that temperatures above optimum for plant growth decreased the carbohydrate reserves.

Loss of TNC in unclipped plants was greatest from anthesis to completion of seed dissemination. This was not anticipated from TNC trends reported in the literature. Typically the lowest level would occur after initiation of spring growth, during the second or third leaf stage. A marked decrease in TNC during seed dissemination also has been reported for orchardgrass (Colby et al., 1966), needle-and-thread (Stipa comata), and squirreltail (Sitanion hystrix) (Coyne and Cook, 1970). In our studies, the process of seed ripening can not entirely account for the TNC decrease, because the TNC of clipped plants decreased similarly to those of unclipped plants. Although the TNC loss in clipped plants may be entirely due to utilization of
reserves following clipping, the similarity of TNC trends of clipped and unclipped plants is striking. The TNC decrease in July is probably not related to temperature because TNC increased during August when the temperature was much higher than during July (Fig. 3).

The decrease in TNC during July was probably related to tiller initiation and growth following the late June and early July rains. White et al. (1972) showed that the percentage of phosphorus and crude protein in the herbage increased during early July indicating renewed growth activity. The weight of stem bases (Fig. 2) increased during the last of July and early August. White et al. (1972) also reported that tiller numbers increased during July, thus indicating that late June and early July rains had caused new tiller initiation. Coyne and Cook (1970) also reported that new tiller growth following seed ripening caused the TNC level to decrease in needle-and-thread, squirreltail, and Indian ricegrass (*Oryzopsis hymenoides*) in Utah.

Loss of photosynthetic area of green needlegrass could have contributed to the TNC decrease during seed ripening. All leaves of floral tillers senesced within one week following seed ripening (White et al., 1972). The first leaf of vegetative tillers initiated during the autumn of 1968 senesced during May 1969. The second, third, and fourth leaves of vegetative tillers initiated in the autumn were 75, 55, and 35 days old, respectively, from the date that their collar appeared. The maximum photosynthetic capacity of the leaves of tall fescue (Jewiss
and Woledge, 1967) and orchardgrass (Treharne et al., 1968) occurred when the leaf blades first emerged from the sheath, and then decreased with age. The life span of an orchardgrass leaf, after it reached full extension, was about 28 days.

The TNC increased during August and early September in both clipped and unclipped plants, even though new leaf length on clipped plants did not exceed 10 cm. During this period the mean air temperature reached 26 C, which is 6.9 C above normal, and the soil water content was below the wilting point in the 0- to 30-cm depth (Fig. 3). Wardlaw (1968) reported that TNC increases if drought restricts elongation growth but does not severely limit photosynthesis. This would seem to be true for both clipped and unclipped green needlegrass plants in late summer.

The resumption of growth after a September 21 rain (Fig. 2) caused the TNC level to decrease during the autumn. McCarty (1938) also found that autumn growth of mountain bromegrass in Utah decreased the carbohydrate reserves.

The seasonal variation of TNC shown in Fig. 4 would not necessarily be characteristic of seasonal variation of TNC in green needlegrass grown at other locations or even for the same location in another year. Colby et al. (1966) showed that the seasonal variation of total fructose in the stem bases of orchardgrass was different when the plants were
grown in Massachusetts than when they were grown in Hokkaido, Japan (Fig. 1).

**Effects of Clipping**

Clipping five times in 1969 reduced yield in 1970 and 1971. While this yield reduction was partially due to the lower TNC level of clipped plants in the autumn of 1969, other factors may be involved. Clipping apparently reduced rooting depth, since we found that less water was extracted from the 90- to 120-cm depth under clipped plants (White and Brown, 1972). Others have found that frequent clipping of orchardgrass essentially stopped root extension (Crider, 1955; Davidson and Milthorpe, 1966b). In other studies, clipping was found to decrease labeled phosphorus uptake for 8 days following clipping (Davidson and Milthorpe, 1966b) or until new roots grew into the labeled phosphorus zone (Oswalt et al., 1959).

In our study, frequent clipping represented an intensity of herbage removal similar to severe grazing. While this treatment reduced yields in subsequent years, it did not kill the plants. Our TNC data show that for unclipped plants, the most critical period for herbage removal was during seed dissemination. Although we did not measure the effects of sequential single clipping on subsequent yield, data from Idaho (Pearson, 1964; Wright, 1967) showed that clipping needle-and-thread grass, a closely related species, during seed ripening
was most detrimental to the following year's herbage production.

Clipping five times in 1969 reduced the TNC level 3 to 5 units at all sampling dates below that of unclipped plants and persisted even after clipping had ceased. Troughton (1957) noted that clipping decreased the TNC level at the end of the growing season proportional to the number of clippings taken during the growing season and inversely proportional to the time interval between clippings. Cook (1966) in a review article noted also that clipping reduced TNC level at the end of the growing season and that the reduction of TNC was inversely proportional to the length of time between the date of the last clipping and the end of the growing season.

Often severe herbage removal by cutting or grazing will decrease herbage production in subsequent years (Weinmann, 1948). Clipping crested wheatgrass grown in arid conditions reduced dry matter yield the following year only when the growing season precipitation was much below normal and it did not decrease yields when the growing season precipitation was much above normal (Hyder and Sneva, 1963). The reduction in dry matter yield was related to reduced carbohydrate reserves present during September. However, clipping perennial ryegrass the previous year had no effect on dry matter production when grown under humid conditions (Baker, 1957; Baker, 1960). The carbohydrate reserves had been restored by early autumn.
Effects of N Fertilization

Environmental conditions play an important role in carbohydrate utilization and storage. Any conditions or treatments, such as higher temperature or N fertilization, that increase meristematic activity with early spring growth should increase the rate and amount of TNC decrease. The greater TNC decrease associated with N fertilization has been documented many times. Rapid depletion of TNC can increase the plant's susceptibility to damage from herbage removal; however, if reserves are used to increase leaf area, this partially offsets the loss of reserves by increasing photosynthetic capacity. The early spring mobilization of energy increases leaf area, herbage productivity, and eventually, TNC concentration. Herbage removal can be delayed to attain these benefits. Following N fertilization, a short delay in herbage removal will allow N to increase the plant's productivity because green needlegrass restores TNC quickly and N fertilization helps restore it even faster.

After the third-leaf stage, N fertilization increased TNC under favorable plant growth conditions and had no effect under unfavorable conditions. Nitrogen increased TNC level during May and June probably because it increased the leaf area. In order to increase the leaf area, N had to increase the leaf width (not measured) because it did not increase the upstretched-leaf height of floral tillers nor the number of leaves per tiller (White et al., 1972). Nitrogen fertilization
increased TNC during seed dissemination because it increased tiller initiation and growth.

During the period of clipping, TNC was sampled at approximately 2 and 13 days after each clipping. The TNC tended to increase between the 2nd and 13th day following clipping in plants on N-fertilized plots. Had TNC been sampled later in the clipping interval (21 days), TNC probably would have been greater in N-fertilized plants. Assuming that these levels at 21 days were equal to or above those at 13 days, TNC must have dropped precipitously during the 2-day period following clipping. The large loss of TNC immediately after clipping has been reported by other investigators (Alberda, 1966; Davidson and Milthorpe, 1966b; Sullivan and Sprague, 1943).

**Total Nitrogen**

Recent reports have suggested that stored protein may be an important source of energy reserves (Davidson and Milthorpe, 1966b; Dilz, 1966; Sheard, 1968). Smith and Silva (1969), however, reported that very little protein was translocated from roots to tops of alfalfa after cutting. Alberda (1966) reported that all dry matter loss in perennial ryegrass plants following a pretreatment -- placing the plants in a nutrient solution in the dark at 30 C for a short period -- could be accounted for by the loss of TNC. In our study, N (crude protein) was not stored and utilized in a manner similar to that of TNC, thus
N is not a major reserve energy source in green needlegrass, which supports Smith and Silva's (1969) conclusion.
SUMMARY AND CONCLUSIONS

The effects of nitrogen (N) fertilization and clipping on seasonal variation of total nonstructural carbohydrate (TNC) concentration in the stem bases of dryland green needlegrass were measured in 1968 and 1969. Residual effects of these treatments were measured in 1970 and 1971. A factorial combination of three levels of N and two levels of clipping were assigned to plots in a completely randomized split-plot design with four replications. Ammonium nitrate was broadcast on main plots in November 1968 at 0, 70, and 140 kg of N/ha. The two clipping treatments applied on the subplots were (A) plants undisturbed until sampled (unclipped) and (B) plants clipped to a 5-cm height at about 21-day intervals on May 7 and 28, June 17, July 8 and 29.

A preliminary study was conducted during October 1968 to determine where green needlegrass stored the major proportion of TNC and number of samples required to adequately measure the percentage of TNC. The preliminary study showed that live stem bases of green needlegrass (October 1968) contained twice the percentage of TNC as live roots. The coefficient of variation of TNC in live stem bases from individual plants was 20% and in live roots, 37%. The coefficient of variation of TNC was 16.5% in live stem bases and 21.7% in live roots when 10 plants were composited per sample and the main plots were arranged in a completely randomized design with four replications.
During 1969, plants were sampled at about 10-day intervals from April 8 through August 12, on September 2 and 27, and on November 8. Stem bases were dried weighed, then analyzed for TNC and total N content. Residual effects of N fertilization and clipping treatments on dry matter yield in 1970 and 1971 were determined from samples harvested when seed was ripe.

The concentration of TNC in the stem bases of unclipped green needlegrass varied considerably during the growing season and was related to growth stage. The percentage of TNC in non-fertilized, unclipped plants decreased from 18 to 14% from growth initiation after winter-dormancy (April 5) until after second leaf formation (April 28). The TNC increased after second leaf formation to 19% just before boot stage (May 26), decreased slightly at heading (May 29) and increased back to 18% at first anthesis (June 9). Following first anthesis, the initiation and growth of new tillers decreased TNC to 9% by completion of seed dissemination (July 26). The TNC increased to 19% in late September during a period in which drought had stopped plant growth. Plant growth resumed after a late September rain and the TNC decreased to 16% by November.

Seasonal variation of TNC in clipped plants was similar to that in unclipped plants but 3 to 5 units lower at all sampling dates. The TNC of clipped plants decreased to a low of 9% on July 10, which was 12 days earlier than the lowest TNC level attained by unclipped plants.
The TNC of clipped plants increased similarly to unclipped plants after July 22. The TNC concentration in clipped plants during November was 13% in contrast to 17% in unclipped plants. The effect of clipping appeared to persist in 1970 and 1971. Clipping significantly decreased dry matter yield in 1970 and appeared to decrease dry matter yield in 1971 (not significant).

Nitrogen fertilization decreased TNC only from growth initiation until the second leaf formation. Thereafter, N fertilization of unclipped plants increased TNC under favorable plant growth conditions and had no effect under unfavorable conditions. Nitrogen fertilization increased the percentage of TNC at heading (May 29) and at first anthesis (June 9). The magnitude of TNC decrease after anthesis was inversely proportional to N fertilization rate and was apparently related to new tiller development and increased photosynthesis following the late June and early July rains. The TNC level of clipped plants on all N treatments decreased in the stem bases of green needlegrass following autumn growth which began after the September 21 rain. The decrease in TNC was greater in plants not fertilized or fertilized with 70 kg of N than in plants fertilized with 140 kg of N. Nitrogen fertilization of clipped plants did not affect TNC within two days after clipping, but increased it within 13 days after clipping. Residual N from the November 1968 application increased dry matter yield of clipped and unclipped green needlegrass in 1970 and 1971.
In the stem bases of unclipped green needlegrass, the percentage of total N varied inversely with the percentage of TNC during the spring and early summer. During the remainder of the season, total N concentrations remained near 1.5% while TNC concentrations fluctuated greatly.
REFERENCES


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Carbohydrate reserves of green needlegrass