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CARBOHYDRATE RESERVES OF BLUE GRAMA (*BOUTELOUA GRACILIS*) AT VARIOUS ENVIRONMENTAL FACTORS UNDER GROWTH CHAMBER EXPERIMENTS

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ABSTRACT

Sods of blue grama (*Bouteloua gracilis*) obtained from the Pawnee Site were used in experiments to determine various plant functions under controlled laboratory conditions that approximated field conditions at the Experimentally Stressed Area (ESA). Three temperature regimes (29.5/18°C, 24/13°C, and 13/7°C day/night schedules) were maintained throughout the experiment. A 12/12 photoperiod was maintained for all experiments. Tops, crowns, and roots were collected and compared among four treatments (fertilized, irrigated, irrigated plus fertilized, and control) for a growing period of approximately 105 days. The dynamics of growth are described using labile and nonlabile plant components during this interval.

INTRODUCTION

Results obtained when blue grama (*Bouteloua gracilis* (H.B.K.) Lag.) plants were exposed to various temperatures and water stressed conditions in growth chambers at NREL during 1971-1972 indicated fluctuations in total available carbohydrate (TAC) and nitrogen and phosphorus contents in the shoots of these plants (Bokhari and Dyer 1973). Generally an increasing trend in all these constituents was observed up to peak growth, followed by a decline as the plants approached maturity. No attempt was made to follow this trend of carbohydrate fluctuations in roots or crowns. The present study was undertaken to investigate the translocation of carbohydrates (flow of energy) from tops to crowns to roots under the influence of differential temperature and water stressed conditions. There is enough information available in the literature on the phenomenon of carbohydrate accumulation in the belowground parts of plants and its subsequent utilization for top growth when new growth is initiated. The latter is not fully understood. When conditions promote rapid growth, reserve carbohydrates, mostly water-soluble, may either decline or remain at low level. In grasses growing under natural conditions the role of reserve carbohydrates is very important in continued functioning of the various components at various trophic levels. As pointed out by White (1973), "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." The effect of various environmental factors and herbage removal

treatments on various aspects of carbohydrate reserves of grasses, namely time of storage, organs of storage, mobilization, and utilization of reserves from these organs for top growth, is not fully understood.

For detailed discussion on the function of carbohydrate reserves in grasses, the reader is referred to the reviews by Graber et al. (1927), May (1960), Cook (1966), McIlroy (1967), and White (1973).

Graber et al. (1927) defined reserve constituents: "Those carbohydrates and nitrogen compounds elaborated, stored and utilized by plant itself as food for maintenance and for the development of future top and root growth." Most investigators have considered carbohydrates as reserves; however, recent studies (Davidson and Milthorpe 1966) have included nitrogenous compounds also as reserves. In a complex and integrated grassland ecosystem not only carbohydrates, but the nutrients especially nitrogen and phosphorus should be included as reserves. The latter in turn would have a profound effect on carbohydrate reserves and finally on the phenomenon of new growth initiation in roots and tops.

The principal carbohydrate reserves in grasses are sugars, fructosans, and starch. Grasses native to cool, temperate climates accumulate fructosans while warm season species accumulate sugars and starch as reserves. Cellulose and pentosans are not considered reserves since they are mainly structural material, they do not exhibit frequent fluctuations in concentration, and they cannot be further utilized in the same way as carbohydrate reserves.

Marshall and Sagar (1965), working with Italian ryegrass, found that labeled CO_2 was not mobilized from roots to the shoots following defoliation, nor was it mobilized from shoots to roots when tops were removed, thus indicating no significant role of root reserves for top regrowth.

Seasonal fluctuations in reserves result from the influence of various environmental factors such as temperature, availability of water, and nutrients. The accumulation of reserves in plants is a dynamic phenomenon which depends upon the balance between photosynthesis and respiration. The level of reserve is determined by growth rate, plant development stage (Hyder and Sneva 1959), and environment (Troughton 1957).

Factors such as nutrients, temperature, water, solar radiation, and photoperiod which determine the magnitude of the gross energy harvested by the primary producers have a direct influence on the energy ultimately available for consumers and decomposers. There also appears to be a significant synergistic interaction between the energy reserves and the nutrient status of the plant. The extent of recycling of the latter would in turn greatly influence the extent of the energy balance in the system and vice versa (Hutchinson 1971). Taking into consideration the whole complex of grassland ecosystem, the survival and continuity which appear to be directly related and dependent on the energy storage of the primary producers, the carbohydrate reserves in roots and crowns deserve special considerations. Any grassland ecosystem model without the input of energy from the belowground biomass may not reflect time adequately. Conditions which determine the energy surplus (the difference between photosynthesis and respiration) of the aboveground biomass differ drastically from the conditions which determine the flow of this excess energy from tops to roots, from roots to soil, and from roots to tops for initiation of new top growth.

Diurnal fluctuations of reserves seem to be minor, though not insignificant in terms of growth. The structure and function of the grassland ecosystem, especially that of a stable one, would be largely dependent on the seasonal energy balance for continuity of this system rather than diurnal fluctuations. At this time, when the effect of consumer and

decomposer trophic levels on the energy flow and nutrient cycling in grassland ecosystem is not fully understood, it is difficult to assume the impact of some of these biotic factors on the energy balance of the primary producer. The energy and nutrient balance of the whole system can be estimated when the contribution of the individual component at each trophic level in terms of energy harvest from the primary producer level is known. The effect of environmental stresses on the energy flow (in terms of carbohydrate equivalents) among various species will be different. Temperature optima for growth and photosynthesis of temperate-origin grasses are less than that of tropical-origin grasses. Similarly the effect of water on reserves varies. Some workers have reported that drought increased the carbohydrate reserves in several grass species (Brown and Blaser 1965; Blaser, Brown, and Bryant 1966); others have reported that drought decreased carbohydrate reserves (Brown 1939; Bukey and Weaver 1939). Under water stressed conditions when increased carbohydrate reserves are reported, there may be transformation of carbon-containing nitrogenous substances (Brown and Blaser 1970). As mentioned earlier, water stressed conditions under extreme temperatures would have a direct effect on photosynthesis and, thus, would markedly reduce the carbohydrate reserves. Again the effect of water and temperature stresses would vary at different development stages. Drastic fluctuations in day/night temperature would have a drastic effect on "reserves." High night temperature would decrease reserves of temperate-origin grasses more than high day temperature, especially in growth chambers.

The effect of nitrogen (N) fertilizer on reserves is variable. The exact role of nutrients on carbohydrate reserves cannot be estimated without considering the availability of nutrients in the soil, the

water potential of the soil, the microbial activities, and the organic matter content of the rhizosphere. Under favorable environmental conditions moderate amounts of N application are believed to have a stimulating effect on carbohydrate reserves. Increased photosynthesis upon N application was reported by Murata (1969). High amounts of N application have been reported to result in reduced carbohydrate reserves. Prianishnikov (1951) interpreted this to be because of the stimulating effect of N on amino acids and amide synthesis, thus reducing the carbon-containing skeleton for carbohydrate reserves.

Many workers have reported the effect of defoliation or grazing on carbohydrate reserves (May 1960). The carbohydrate reserve level in the roots and tops depends upon the time and frequency of cutting, the species, and the environmental factors. Frequent and intensive grazing or cutting reduces the reserves in the root. Herbage removal reduces the amount of carbohydrate reserves, root growth, and leaf area (Alcock 1964). The importance of reserves in controlling regrowth after herbage removal is a controversial topic. Some scientists (e.g., May 1960) believe that the role of reserves in initiating new growth or in determining the rate of regrowth has not been firmly established while others (Ehara, Maeno, and Yamada 1966; Pearce, Fissel, and Carlson 1969; Smith and Marten 1970) have reported that reserves are definitely used for regrowth following herbage removal.

Thus the importance of carbohydrate reserves in controlling regrowth following herbage removal is not fully understood and is still a topical subject in grassland management. A critical level of carbohydrate reserves for regrowth has not been determined in many grasses. Blue grama grown in a growth chamber (at the Natural Resource Ecology Laboratory) was

unable to initiate regrowth at carbohydrate reserve levels in roots and crowns below 2.5% and 3.5%, respectively. However, when moderate amounts of N (30 kg N/ha) were applied to the same plants, growth was initiated within 2 weeks. From this it appears that merely the presence of carbohydrate reserves in roots or crowns does not determine the rate of regrowth when other factors are limiting. In this case failure of regrowth initiation by carbohydrate reserves in the presence of adequate amounts of water (at field capacity) and in the absence of N suggests that the phenomenon of carbohydrate mobilization may be independent of the level of carbohydrate reserves. It appears that utilization of carbohydrate reserves depends upon the availability of a factor which is synthesized in the presence of adequate amounts of N. The synthesis of this factor in turn may depend upon the availability of energy that is supplied by the carbohydrate reserves. Another alternate interpretation of the effect of moderate nitrogen applications on carbohydrate mobilization or regrowth may be that, in fact, N application has no effect on mobilization of carbohydrate per se, but rather on the tiller primordium where new growth initiation is intended.

We suggest that more research is needed to establish the role of a critical amount of carbohydrate reserves for regrowth in various grass species in conjunction with other factors. The triggering mechanism, if it really exists, will vary from species to species, and its role or mode of action may be different at different times during and after plant growth.

Those grassland management practices, which emphasize carbohydrate reserves only, may not turn out to be so important, especially when theoretically adequate amounts of carbohydrate reserves fail to initiate regrowth.

In the natural grassland ecosystem the interrelated and integrated effects of various biotic and abiotic factors on energy balance and nutrient cycling seem to be inseparable, and if the system is operating at a steady state, minor external disturbances in terms of environmental factors are adjusted in favor of stabilization of the system.

In this study blue grama plants were subjected to various temperature regimes under four treatments in growth chambers. The objective of the study was to investigate the influence of temperature, water stress, and nitrogen fertilizer and their interaction effect on the carbohydrate reserves in aboveground and belowground biomass at various phenological stages. Both labile and nonlabile carbohydrate components are considered.

METHODS AND MATERIALS

Blue grama plants growing in one of the campus greenhouses were brought to the Natural Resource Ecology Laboratory and were separated into individual sods. Only "healthy-looking" roots and leaves were used. They were transferred to plastic pots (12.7 cm deep × 12.7 cm surface diameter) containing a soil potting mixture consisting of soil from Pawnee Site, sand, and peat moss (4:1:1). The Pawnee Site soil representing various treatment areas was heterogenous in texture and fertility levels. The soils were thoroughly mixed before being combined with sand and peat moss. A total of 120 pots were filled with this soil mixture and placed in the three growth chambers in groups of 40. These were watered to field capacity to bring the water content to uniform level before planting. A week later plants were planted in each pot and left in the same chamber for another 2 weeks to precondition them to the chamber environments. Treatments began at the end of the 2-week preconditioning period. Out of the 40 pots, 10 received no treatment (control = C), 10 were given nitrogen

(N) at the rate of 150 kg N/ha in the form of ammonium nitrate, 10 received water daily to maintain field capacity, and the remaining 10 were given nitrogen and a daily watering. The control and the fertilized pots received water at 3- to 4-day intervals to prevent them from extreme drought conditions.

Environmental conditions in the three growth chambers were 13/7°, 24/13°, and 29.5/18°C day/night temperature, alternating with 12-hour photoperiods at 2000 ft-c light intensity.

Sampling was started a week later, following the nitrogen application (0 day). Subsequent samples were taken on the 45th, 65th, 90th, and 120th day counting from 0 day.

At each sampling time eight pots from each chamber (two from each treatment) were taken; separated into shoots, crowns, and roots; dried at 70°C for 48 hours; and held for chemical analyses. Roots along with the adhering soil particles were first dried and then separated by hand from the soil. Dry weight of each plant part was recorded after drying and ground through 40-mesh screen in a Wiley mill.

A 500-mg sample was used for determining TAC by the Smith (1969) method using 0.2 N H_2SO_4 to hydrolyze the starches.

RESULTS AND DISCUSSION

The time-course responses of dry weight of plants from the four treatments at different temperature regimes are given in Fig. 1, 3, and 5.

The magnitude of the range of fluctuations in carbohydrate reserves of the three plant parts are given in Fig. 2, 4, and 6 while Fig. 7 to 9 show the rate of growth over a period of time. An analysis of variance was made to determine the significance of differences in the growth, yield, and

carbohydrate reserves because of temperature, water stress, fertilization, time, and all interactions among these factors. All the differences among these interactions were highly significant ($p<0.01$) (Tables 1 to 3).

In comparing Fig. 1, 3, and 5 the data from four treatments reveal two aspects of plant growth quite clearly: (i) irrigated plus fertilized plants produced significantly more total yield under the three temperature regimes than did any of the plants under the remaining three treatments, and (ii) growth rate of plants that received water plus fertilizer was faster at 29.5/18°C than under 13/7°C or 24/13°C.

Plants that received only water treatment recorded higher growth rates than the control or the fertilized plants. The latter had the slowest rate of growth.

Response to 13/7°C Temperatures

Dry matter production. The dry weight of shoots, crowns, and roots of control plants under 13/7°C increased from 3.20, 2.32, and 1.34 g (0 day) to 8.30, 3.42, and 2.67 g in 120 days, an increase of 160%, 47%, and 100%, respectively (Fig. 1). During the same period the dry weight of the same three organs of plants from fertilized treatment increased from 3.51, 2.15, and 1.27 g to 8.71, 3.47, and 2.58 g, an increase of 148%, 61%, and 103%, respectively. Irrigated plants produced significantly greater yields than the control or the fertilized plants. Increases of 200%, 80%, and 117% were recorded in 120 days for shoots, crowns, and roots, respectively. The irrigated plus fertilized plants accumulated more dry matter than the rest of the plants. The shoot, crown, and root increases were 226%, 112%, and 138%, respectively.

Table 1. ANOVA[a/] of dry weight/pot.

Source	df	SS	MS	F	
Treatment (Trt)	3	265.66	88.55	5903.33	***[b/]
Temperature (Temp)	2	61.60	30.80	2053.33	***
Trt × Temp	6	24.67	4.11	274.00	***
Error 1	12	0.18	0.015		
Days (D)	4	740.59	185.15	12879.30	***
D × Trt	12	80.06	6.67	464.00	***
D × Temp	8	18.94	2.37	164.87	***
D × Trt × Temp	24	10.71	0.45	31.30	***
Error 2	48	0.69	0.014		
Position (P)	2	2035.07	1017.53	39388.26	***
P × Trt	6	135.63	22.60	874.84	***
P × Temp	4	14.91	3.73	144.39	***
P × Trt × Temp	12	23.36	1.95	75.48	***
Error 3	24	0.62	0.026		
Days × Position	8	355.46	44.43	5017.98	***
D × P × Trt	24	43.66	1.82	205.55	***
D × P × Temp	16	9.15	0.57	64.38	***
D × P × Trt × Temp	48	13.50	0.28	31.62	***
Error 4	96	0.85	0.009		
Total	359	3835.33			

[a/] Analysis of variance.

[b/] ***: Significant for $\alpha = 0.01$.

Table 2. ANOVA[a/] of % TAC.

Source	df	SS	MS	F	
Treatment (Trt)	3	669.43	223.14	1369.66	***[b/]
Temperature (Temp)	2	82.17	41.08	252.15	***
Trt × Temp	6	39.81	6.63	40.70	***
Error 1	12	1.955	0.163		
Days (D)	4	696.44	174.11	1313.62	***
D × Trt	12	189.65	15.80	119.21	***
D × Temp	8	20.87	2.61	19.69	***
D × Trt × Temp	24	22.24	0.93	7.02	***
Error 2	48	6.362	0.133		
Position (P)	2	5628.36	2814.18	21212.41	***
P × Trt	6	383.56	63.93	481.88	***
P × Temp	4	42.96	10.74	80.95	***
P × Trt × Temp	12	21.64	1.80	13.57	***
Error 3	24	3.185	0.133		
Days × Position	8	422.47	52.81	383.38	***
D × P × Trt	24	142.32	5.93	43.05	***
D × P × Temp	16	16.19	1.01	7.33	***
D × P × Trt × Temp	48	23.91	0.50	3.63	***
Error 4	96	13.22	0.138		
Total	359	8426.74			

[a/] Analysis of variance.

[b/] ***: Significant for $\alpha = 0.01$.

Table 3. ANOVA[a/] of growth rate.

Source	df	SS	MS	F	
Treatment (Trt)	3	36715.36	12238.45	862.12	***[b/]
Temperature (Temp)	2	7985.72	3992.86	281.27	***
Trt × Temp	6	4739.64	789.94	55.65	***
Error 1	12	170.35	14.20		
Days (D)	3	18178.29	6059.43	145.18	***
D × Trt	9	4381.71	486.86	11.67	***
D × Temp	6	3537.42	589.57	14.13	***
D × Trt × Temp	18	2991.53	166.20	3.98	***
Error 2	36	1502.51	41.74		
Position (P)	2	152891.56	76445.78	9664.45	***
P × Trt	6	18438.52	3073.09	388.51	***
P × Temp	4	3534.09	883.52	111.70	***
P × Trt × Temp	12	6203.12	516.93	65.35	***
Error 3	24	189.84	7.91		
Days × Position	6	28731.95	4788.66	165.00	***
D × P × Trt	18	11908.74	661.60	22.80	***
D × P × Temp	12	3838.40	319.87	11.02	***
D × P × Trt × Temp	36	10461.06	290.58	10.01	***
Error 4	72	2089.55	29.02		
Total	359				

[a/] Analysis of variance.

[b/] ***: Significant for $\alpha = 0.01$.

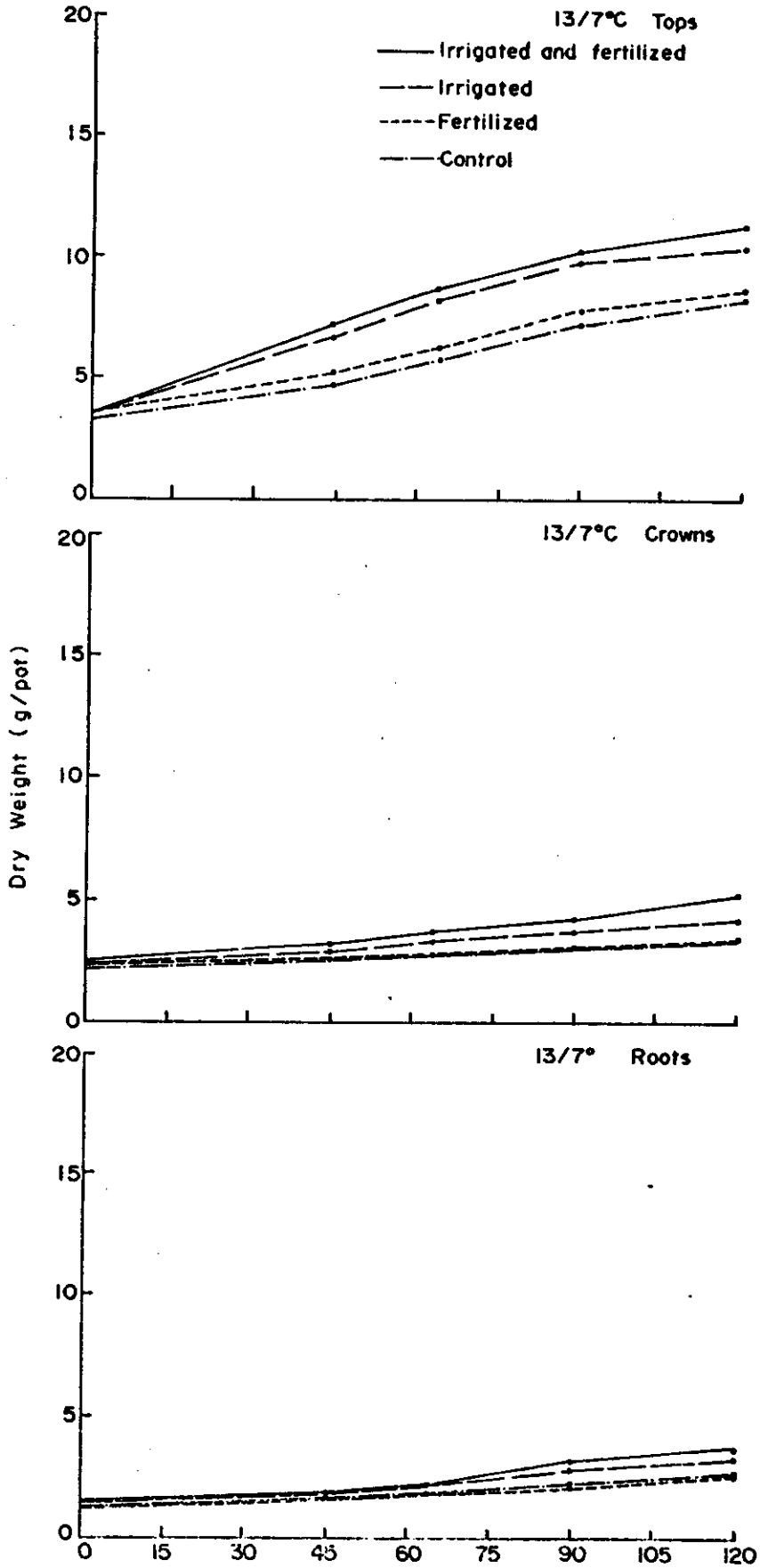

Fig. 1. Dry weight of shoots, crowns, and roots at 13/7°C.

These results indicate that if other factors are not limiting, then blue grama plants were able to utilize the added nitrogen and water more efficiently. Since fertilization in the absence of adequate water failed to increase the yield of blue grama, it appeared that blue grama plants in the presence of adequate water had developed the capability to utilize this added nitrogen for the production of more yield. Added nitrogen and water enhanced shoot growth and leaf area and, thus, increased the photosynthetic activities of shoots. Increase in shoot production was followed by increases in crowns and roots.

Total available carbohydrates (TAC). Fig. 2 gives the carbohydrate reserves at 13/7°C from four treatments. The TAC of irrigated plus fertilized shoots, crowns, and roots increased from 6.75% to 18.32% (in 90 days), from 2.52% to 5.25%, and from 1.65% to 3.75% (in 120 days), respectively. However, the TAC of shoots dropped from 18.32% to 13.20% in 30 days following the 90-day growth period (Fig. 2). A slight decrease in TAC of roots was also recorded. Similarly the TAC of irrigated shoots increased from 6.85% to 15.75% in 90 days, followed by decreases that dropped to 11.30% at the end of 120 days.

The TAC of crowns and roots at the same time increased from 2.45% to 4.87% and from 1.52% to a maximum of 3.20%, respectively. The latter dropped to 2.85% in the next 30-day growing period. The TAC of fertilized and untreated plant shoots increased from 6.35% to 9.25% and from 6.45% to 10.85% in 90 days, respectively, followed by slight decreases towards the end of the growing season. The TAC of crowns and roots of the same plants showed slight increases as the plants approached maturity.

When compared with the dry matter yield of these plants under 13/7°C, an inverse relationship appears to exist between TAC and dry matter production.

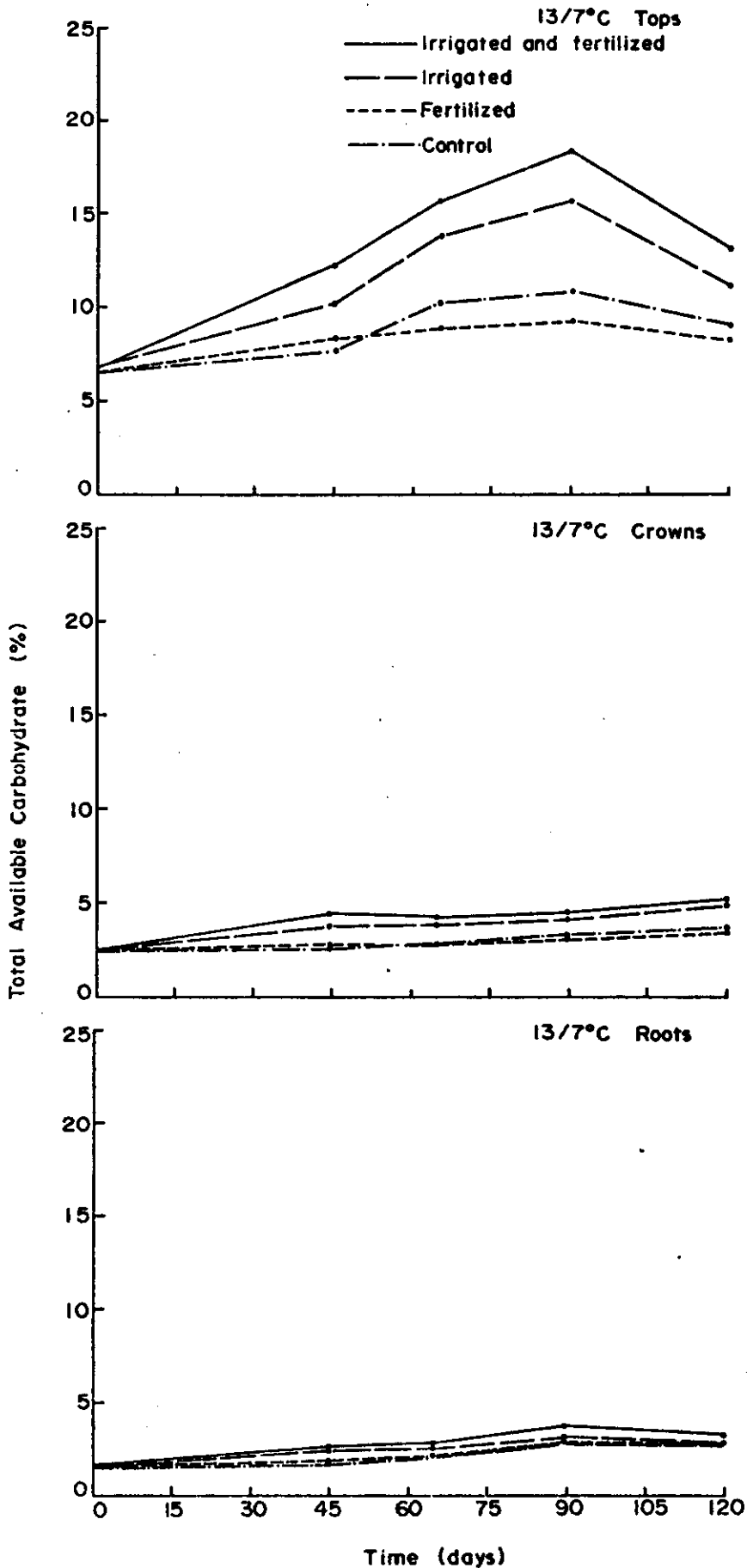

Fig. 2. Total available carbohydrates at 13/7°C.

The TAC in shoots of plants under the four treatments developed a tendency of decreasing trend toward the end of growth period; however, there was no significant change in the TAC of belowground parts during the same interval. This indicates that either there is more translocation of carbohydrate reserves to belowground parts or production of more fibrous materials as the plant approaches maturity. The continuous increase in dry matter production and at the same time a similar increase in TAC of belowground parts suggest that both translocation and dilution phenomena occur simultaneously. The significantly increased TAC contents in shoots, crowns, and roots of irrigated plus fertilized plants further support these assumptions with respect to dry matter production under the same treatment. For blue grama plants added nitrogen and water have a very favorable effect on growth.

Response to 24/13°C Temperatures

Dry matter. In Fig. 3 the results at 24/13°C are given. The dry weight of shoots, crowns, and roots of plants under the four treatment conditions exhibited greater increases over the plants from the same treatment at 13/7°C. Control plants increased 222%, 60%, and 124%, and fertilized plants increased 157%, 62%, and 135% for the respective plant parts.

The shoot dry weight values of the untreated plants were higher than the fertilized plants while the reverse was true for crowns and roots. At the same temperature regimes the plant dry weight values for the irrigated and the irrigated plus fertilized plants were much greater than for untreated and fertilized plants.

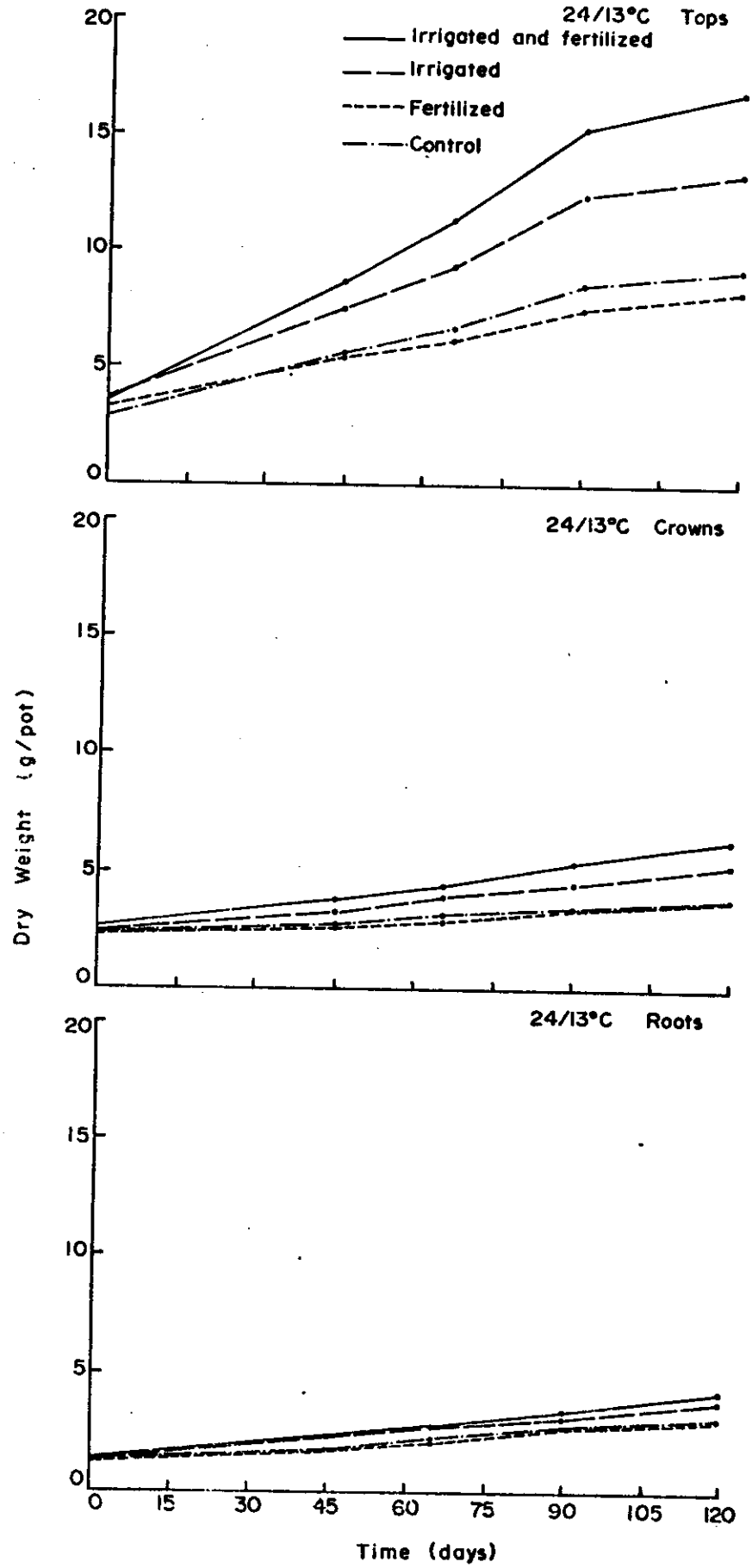

Fig. 3. Dry weight of shoots, crowns, and roots at 24/13°C.

The irrigated and the irrigated plus fertilized plant increases in dry matter production for the three plant parts were 270%, 112%, and 183% and 378%, 138%, and 194%, respectively, for shoots, crowns, and roots. Irrigated plus fertilized plants were more efficient in dry matter accumulation than irrigated plants.

Total available carbohydrates (TAC). In Fig. 4 the TAC at 24/13°C from the four treatment groups is given. In general, the TAC of shoots, crowns, and roots at 24/13°C is greater than at 13/7°C (Fig. 2 and 4). The shoots of irrigated plus fertilized plants increased in TAC contents from 7.23% to 20.20% (in 90 days), dropping to 15.32% within the next 30 days. The TAC of crowns and roots showed a gradual increase until the end of the growing period with a slight decrease in TAC of roots following the 90-day growth period.

On the other hand, the TAC component of irrigated shoots increased from 7.23% to 17.25% (in 90 days), dropping to 13.75% toward the end of experimental period. The TAC of crowns and roots showed gradual increases, reaching a maximum of 6.35% in the case of crowns and 4.15% in the case of roots in 120 days.

The TAC contents of shoots, crowns, and roots from untreated and fertilized plants were of significantly lower magnitude than those of the other two treatment groups.

The TAC of untreated and fertilized plant shoots increased from 6.69% to 11.87% and 7.12% to 10.83% in 90 days, followed by decreases within the next 30 days and dropping to 8.75% and 7.83%, respectively.

The TAC of crowns and roots of untreated plants followed gradual increases from 2.48% to 3.55% and 1.50% to 2.75%, respectively.

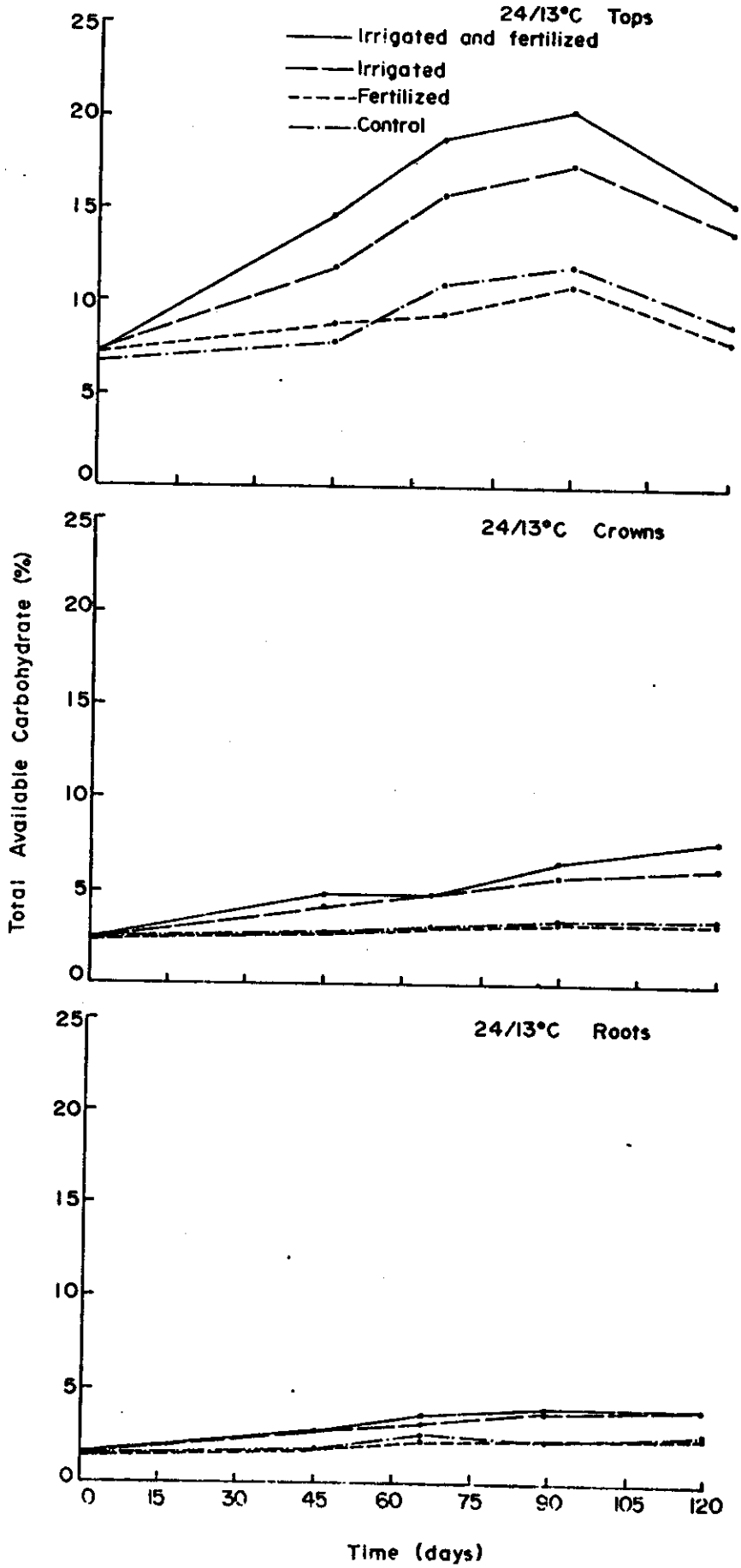

Fig. 4. Total available carbohydrates at 24/13°C.

Similarly for crowns and roots of fertilized plants, increases of 2.47% to 3.42% and 1.50% to 2.58%, respectively, were recorded.

Here also the irrigated plus fertilized plants under 24/13°C were able to produce more yield and TAC than the plants from the other three treatments. Plants from all four treatments under 24/13°C exhibited significantly greater dry matter production and TAC content in shoots, crowns, and roots than the plants under 13/7°C. It appears that at higher temperature regimes blue grama plants are able to utilize the added nitrogen and water more efficiently. More carbohydrate reserves were accumulated in crowns and roots under 24/13°C than under 13/7°C.

Response to 29.5/18°C Temperatures

Dry matter production. In Fig. 5 the results at 29.5/18°C for the plants from the four treatments are given. Here the dry matter production in all three parts of the plant was greater than for the two previously mentioned temperature regimes. Dry matter of the shoots, crowns, and roots of untreated plants increased 210%, 56%, and 106%, respectively, while fertilized plants at the same time showed increases of 150%, 51%, and 110%. Again the shoots and crowns of untreated plants had accumulated more dry matter than did the fertilized plants.

The increases in the weight of shoots, crowns, and roots from the irrigated plants were 317%, 127%, and 245% and for the irrigated plus fertilized plants were 372%, 148%, and 243%.

Total available carbohydrates (TAC). In Fig. 6 the TAC content at 29.5/18°C is given. When the results in this figure are compared with the dry weight results in Fig. 5, an inverse relationship between the dry matter

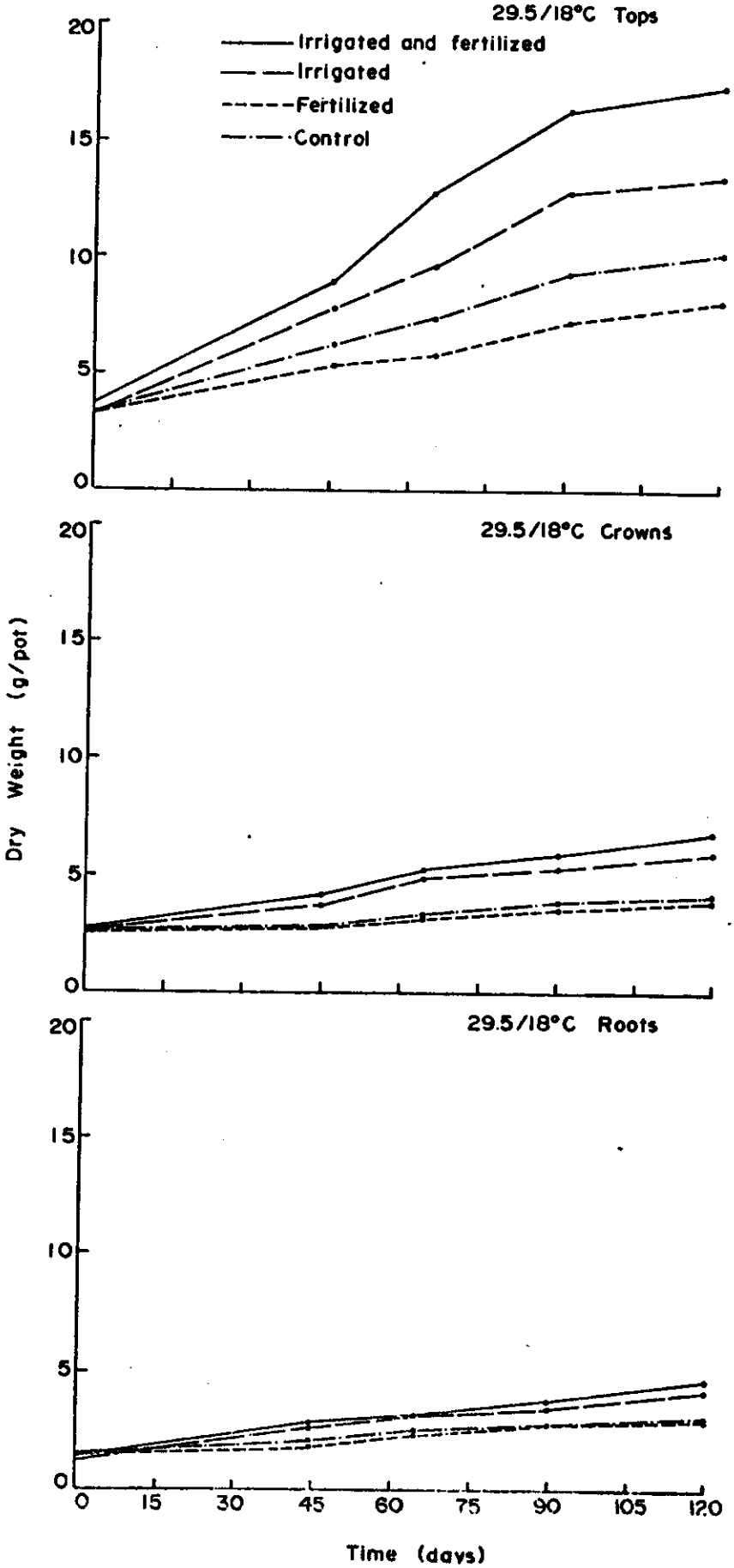

Fig. 5. Dry weight of shoots, crowns, and roots at 29.5/18°C.

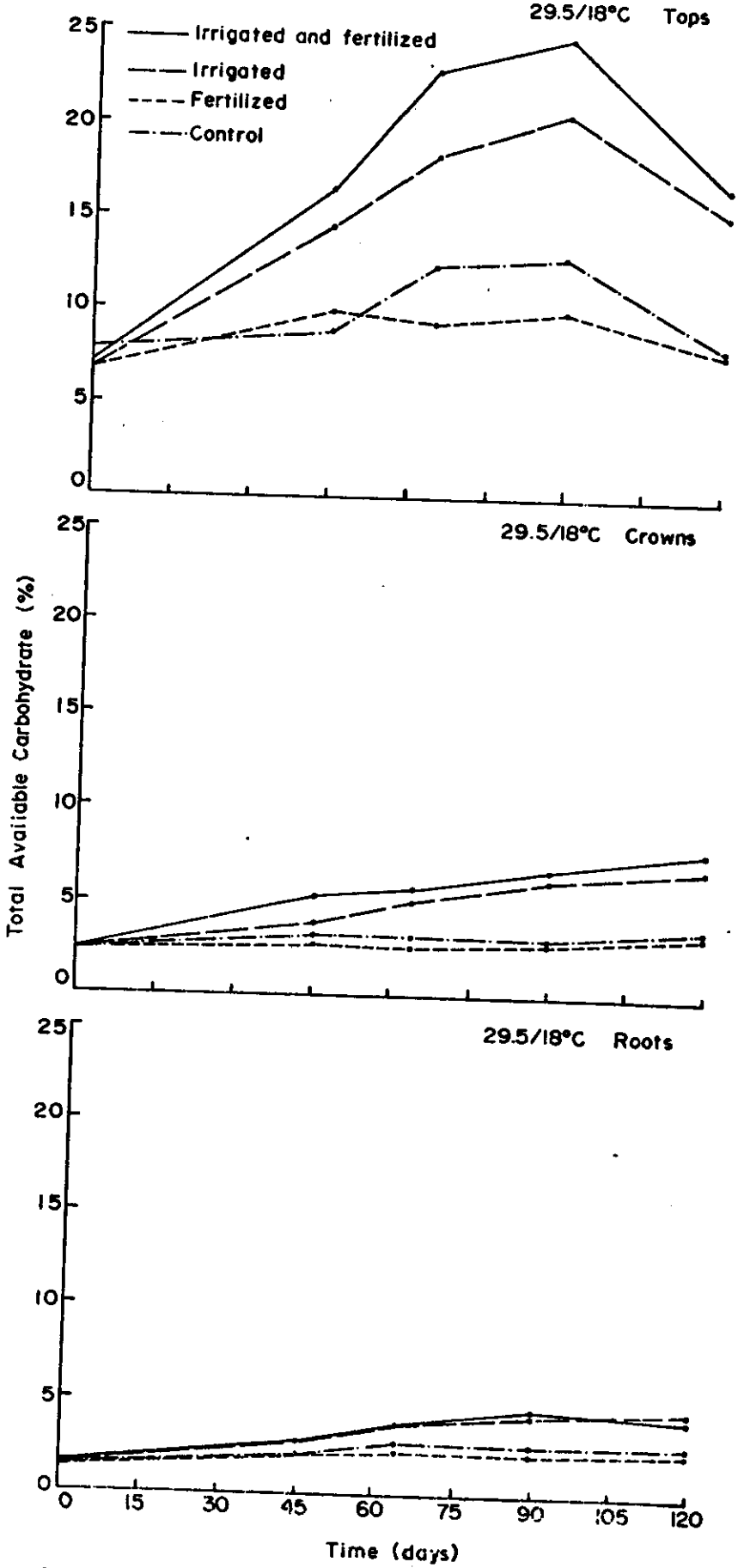

Fig. 6. Total available carbohydrates at 29.5/18°C.

production and the carbohydrate reserves becomes evident toward the end of the experimental period. This relationship is also true for the other two temperature regimes.

The TAC content of shoots from fertilized plus irrigated plants increased from 7.12% to a maximum of 24.60% in 90 days and then declined within the next 30 days to 16.81%. The TAC of crowns and roots increased gradually from 2.46% to 7.87% and 1.55% to 4.23%, respectively, in 120 days.

The irrigated plant shoots exhibited a similar trend which showed increases in TAC content from 6.81% to 20.50% in 90 days, dropping to 15.25% at the end of 120-day growth period. The TAC of crowns and roots exhibited a similar trend as that of irrigated plus fertilized plants but with a somewhat lesser magnitude.

The TAC content of fertilized and untreated plants was significantly lower than the other two treatment groups (Fig. 6). Fertilized and untreated plant shoots reached a maximum of 9.87% and 12.84% TAC, respectively, in 90 days. The TAC contents of crowns and roots from the same plants were 3.38% and 2.43% (fertilized) and 3.67% and 2.87% (untreated), respectively, at the end of 120 days.

These results indicate very clearly that the most favorable response of blue grama plants to added nitrogen and water are obtained at high temperature regimes. Blue grama is a warm season species, and thus such a response would be expected under the conditions of this experiment. The response of many grasses to added nitrogen is quite controversial (White 1973). Favorable response to nitrogen under conditions of deficient soil nitrogen has been reported (Murata 1969). High amounts of added nitrogen were reported by Prianishnikov (1951) to reduce carbohydrate reserves.

In this study added nitrogen in the presence of adequate water was found to have a stimulating effect on growth and carbohydrate reserves. The results of this study are not in agreement with the work of Trlica and Cook (1972) who reported that added water reduced carbohydrate reserves in crowns and roots of crested wheatgrass and Russian wild rye. This discrepancy could be caused by several factors. In this experiment blue grama plants were grown under controlled environmental conditions, and as such, factors that might have been limited in the field under adequate soil water were not limited in this study. Blue grama is a warm season species, and thus its response to water and nitrogen might be altogether different than cool season species. Again plants grown under a continuous watering schedule in the presence of adequate nutrients appear to not only enhance the shoot growth, but also increase translocation of carbohydrate reserves to belowground parts.

Growth Rate

Growth rates, expressed in g/pot/day, are given for blue grama plants in three temperature regimes and four treatments (Fig. 7 to 9). Higher growth rates of shoots were obtained by plants in the irrigated and fertilized plus irrigated treatments. Control- and fertilized-plant shoots at 13/7°C obtained a maximum growth rate of 0.06 g/pot/day on the 65th day while irrigated and fertilized plus irrigated plant shoots reached a maximum growth rate of 0.08 g/pot/day at the same time, an increase of 33% (Fig. 7). The crowns and roots from control and fertilized plants reached a maximum of 0.01 g/pot/day, respectively, in 90 days. The maximum growth rate of crowns for irrigated and fertilized plus irrigated plants during the 65-day growth period was 0.03 g/pot/day. However, roots of fertilized plus irrigated plants obtained a higher growth rate (0.03 g/pot/day) than the irrigated

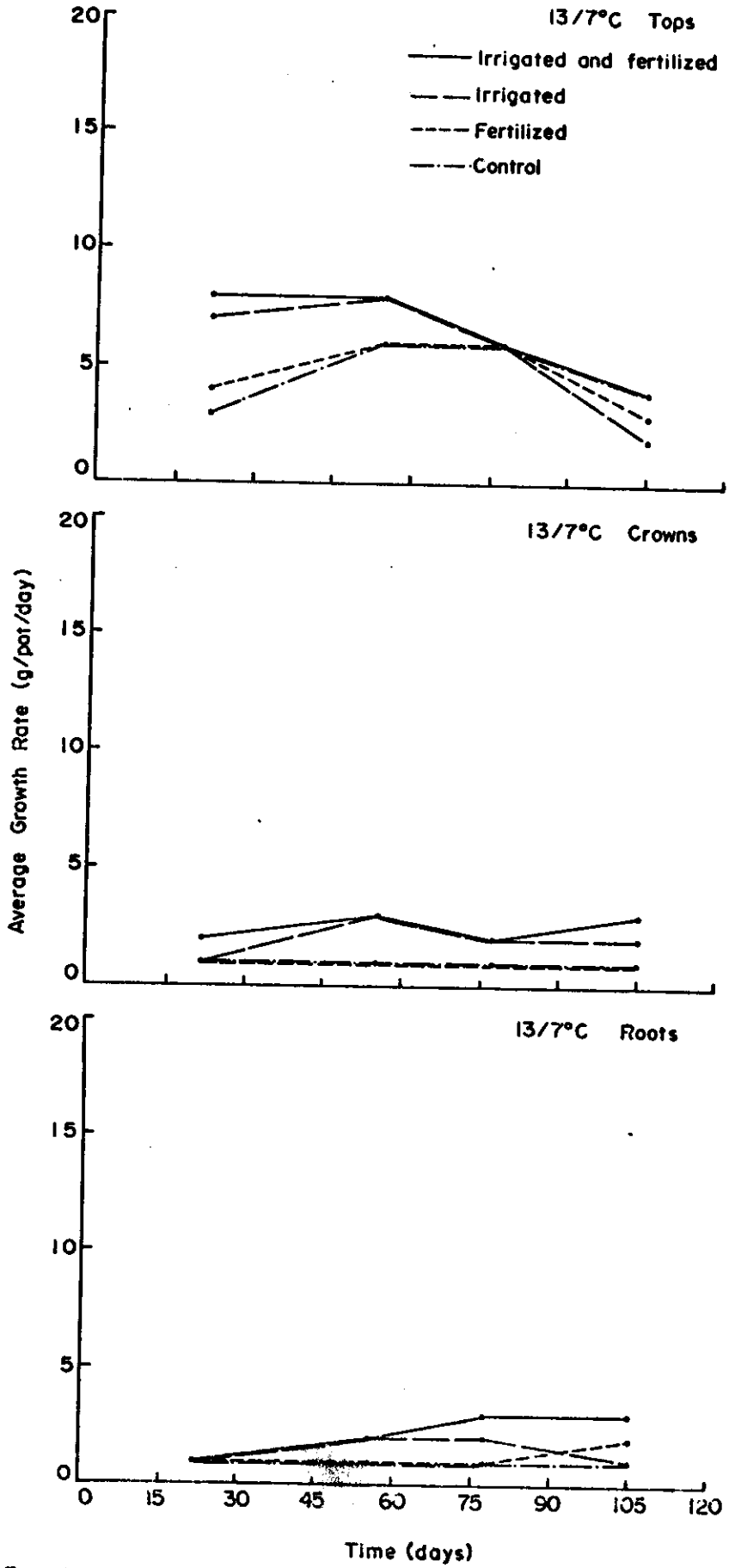

Fig. 7. Growth rate for blue grama plants at 13/7°C.

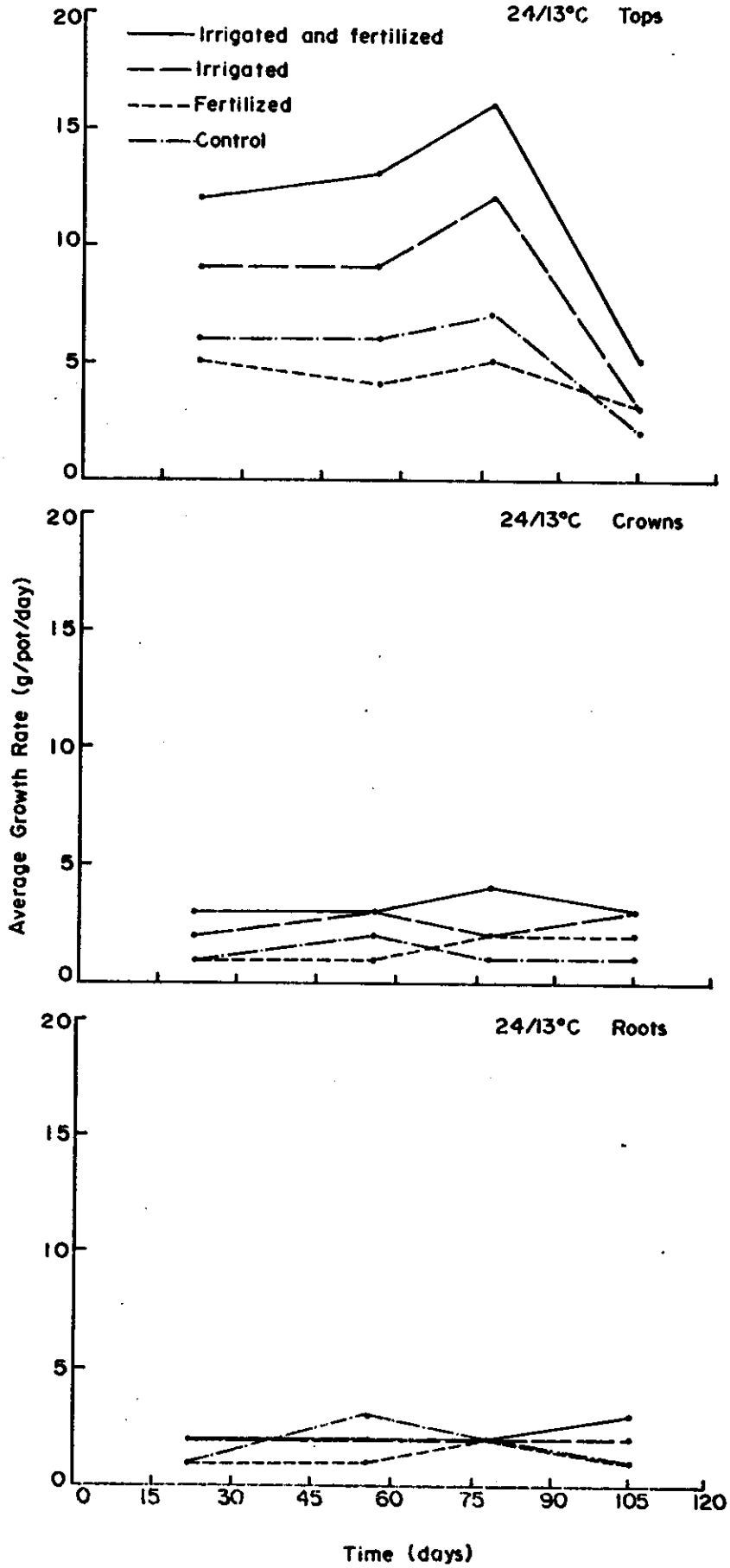

Fig. 8. Growth rate for blue grama plants at 24/13°C.

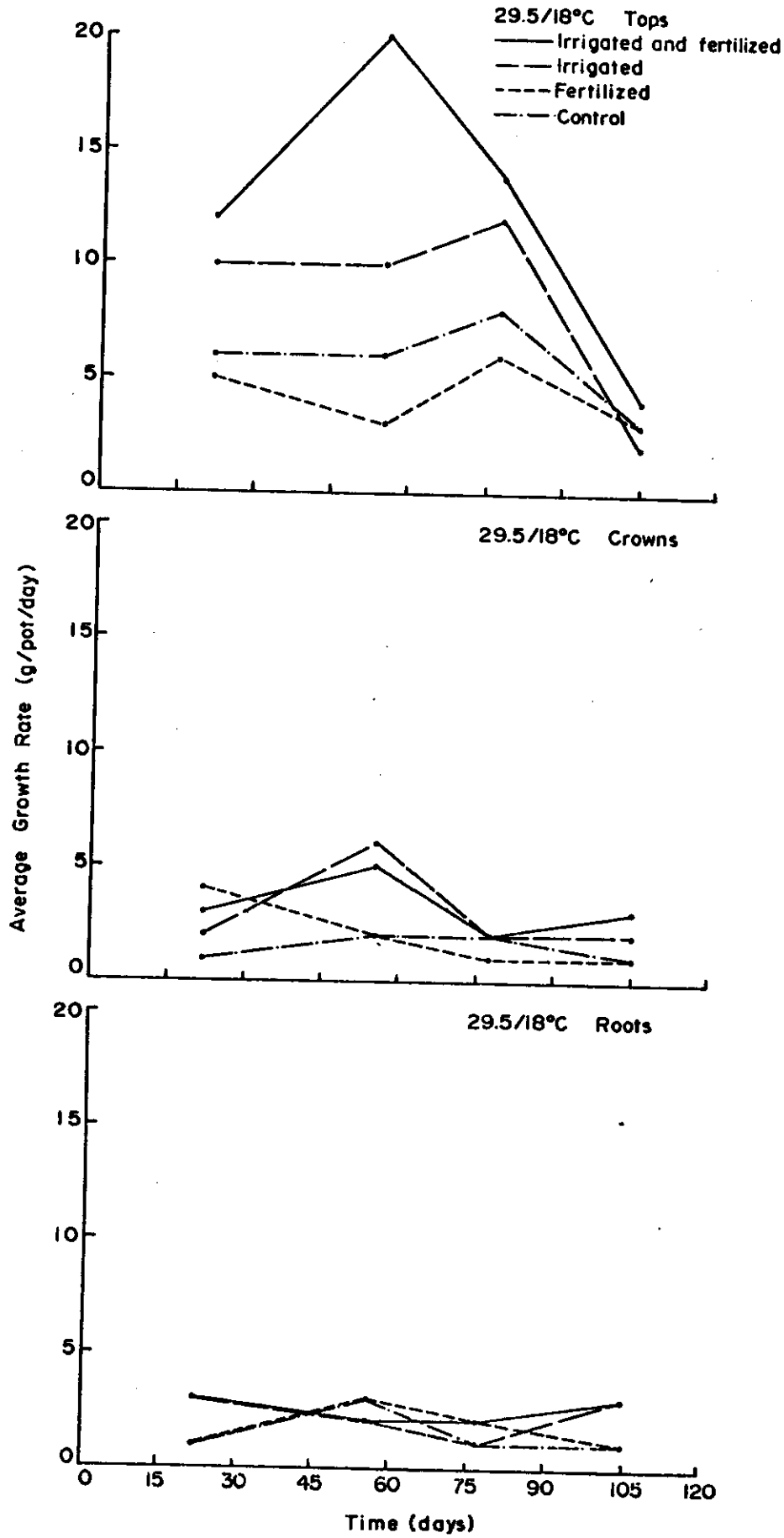

Fig. 9. Growth rate for blue grama plants at 29.5/18°C.

plants (0.02 g/pot/day). At 24/13°C temperature the shoot growth rate under the four treatments was significantly greater at peak growth than for 13/7°C temperature. The shoots of control plants at peak growth (90 days) recorded a growth rate of 0.07 g/pot/day while the fertilized plant shoots reached a maximum growth rate of 0.05 g/pot/day during the same interval, a decrease of 40% (Fig. 8).

The shoots of irrigated plants at this temperature (24/13°C) had a growth rate of 0.12 g/pot/day, and the fertilized plus irrigated plants recorded a growth rate of 0.16 g/pot/day at the same time, an increase of 33%. The maximum growth rates of crowns and roots under the four treatments at this temperature (24/13°C) were nearly the same as for 13/7°C, except for the crowns of fertilized plus irrigated plants where a slightly higher growth rate (0.04 g/pot/day) was recorded.

Under the 29.5/18°C temperature regime, shoot, crown, and root growth rates were higher for all four treatments in contrast to 24/13°C or 13/7°C temperatures. Maximum shoot growth of control, fertilized, irrigated, and fertilized plus irrigated plants at peak growth (90 days) was 0.08, 0.06, 0.12, and 0.14 g/pot/day, respectively. Increases of the same magnitude in growth rate of crowns and roots were also recorded. There was no significant difference in crown and root growth rates between control and fertilized plants. The same was true for irrigated and fertilized plus irrigated crowns and roots.

As evident from Fig. 9, maximum shoot growth under the four treatments was obtained at 29.5/18°C. On the other hand, maximum crown and root growth at 29.5/18°C was recorded only in irrigated and fertilized plus irrigated plants. The crown and root growth rate of control and fertilized plants under the three temperature regimes remained almost the same.

These results indicate that maximum shoot, crown, and root growth rates of blue grama plants can be achieved at temperatures greater than 24/13°C day/night temperatures. Growth rate can be enhanced in the presence of adequate water and nitrogen fertilization. There seems to be a synergistic effect of soil water and nitrogen on plant growth rate. The interaction effects of temperature, water, and nitrogen on plant growth rate are highly significant at high temperature regimes. Added water and nitrogen at low temperature regimes (13/7°C) appear to have no appreciable effect on growth rate. These results in general suggest that high temperatures accompanied by adequate water, soil nutrients, and other nonlimiting factors increase not only growth of plants, but also result in the accumulation of greater amounts of carbohydrate reserves in belowground parts.

CONCLUSION

Variation of carbohydrate reserves on diurnal and seasonal time scales is a common phenomenon in plant tissue maintaining a dynamic system of energy balance (White 1973). The magnitude of energy balance at any time among various parts of plants is dependent on various environmental factors such as temperature, soil water, nutrient status of the plant, and origin of the plants. In this study high temperature regimes and adequate water plus N treatment showed a very favorable effect on TAC of shoots (Fig. 2, 4, and 6). When accompanied by adequate soil water, nitrogen applications under N deficient conditions in the soil appear to enhance growth, even at high temperatures. Being a warm seasen grass, the temperature optima of blue grama are higher than a cool season grass. Under conditions of low soil water N

applications have adverse influence on growth as well as on carbohydrate reserves. According to Prianishnikov (1951), excess nitrogen tends to decrease carbohydrate reserves when other factors do not limit plant growth. He attributes this to the enhanced utilization of carbon compounds for amino acid synthesis. This study does not bear out these previous findings. Here we indicate that an addition of N appears to have no such detrimental effect on carbohydrate reserves or on growth when accompanied by adequate water and suitable temperature conditions. The adverse effect of N on growth and TAC under drought conditions are evident in this study. Blue grama plants do not appear to respond to N under high water stress conditions.

Blue grama plants show an inverse relationship between carbohydrate content and growth as plants approach maturity. This observation is apparent under all three temperature regimes. At a low temperature regime this tendency becomes quite apparent before plant maturity.

The distribution of carbohydrate reserves among the three principal parts of the plants seemed to follow a natural cycle throughout the experimental period with the shoots undergoing significant fluctuations close to peak growth. This was true in all temperature regimes under all four treatments. Carbohydrate reserves in crowns and roots did not exhibit such fluctuations, but rather gradual increases were recorded throughout the experimental period.

The fluctuations of the carbohydrate reserves in shoots plus the gradual increases in crowns and roots indicate that during and after peak growth the excess carbohydrates are translocated to the belowground

parts of plants as food reserves. Another explanation for the decline of TAC in shoots could be the accumulation of more fibrous materials, thus exerting a dilution effect simultaneously. The amount of carbohydrate reserves in the three parts of plants at any given time during the 120-day growth period was greater at high temperature regimes, especially in the fertilized plus irrigated plants. This would suggest enhanced photosynthetic activity in excess of respiration which results in an excess of carbohydrate translocation to belowground parts.

The growth of belowground parts was correlated to its carbohydrate reserves under any given temperature regime, thus indicating a steady energy flow in these parts. However, the magnitude of this balance was greater at high temperature regimes in the fertilized plus irrigated plants followed by irrigated, unirrigated, and fertilized plants in that order.

The dry matter and total available carbohydrates components for shoots, crowns, and roots were converted to energy quanta to ascertain the energy flow levels in plants at various intervals. These data are available in Bokhari, Singh, and Smith (1974). Some of the results are presented here (Appendices) for information that can be used internally in the program. The gross dry weight of plants per pot (Appendix I) was converted to gross energy by multiplying with a factor of 4.3 kcal g^{-1} dry weight. The TAC data (Appendix II) were converted to labile energy by multiplying with a factor of 3760 cal g^{-1} glucose (Appendix III). To obtain tthe nonlabile fraction of energy (Appendix IV), labile energy was subtracted from gross energy. Thus the tables in Appendices III and IV show the magnitude of labile and nonlabile energy in shoots, crowns, and roots of blue grama under various temperatures, water stress, and fertilizer conditions.

LITERATURE CITED

Alcock, M. B. 1964. The physiological significance of defoliation on the subsequent regrowth of grass-clover mixtures and cereals, p. 25-41. *In* D. J. Crisp [ed.]. Grazing in terrestrial and marine environments. Blackwell Sci. Publ., Oxford, England.

Blaser, R. E., R. H. Brown, and H. T. Bryant. 1966. The relationship between carbohydrate accumulation and growth of grasses under different microclimates. Int. Grassland Congr., 10th Proc. (Helsinki, Finland), p. 147-150.

Bokhari, U. G., and M. I. Dyer. 1973. The effects of environmental stresses on growth of blue grama (*Bouteloua gracilis*) in environmental control growth chambers. US/IBP Grassland Biome Tech. Rep. No. 227. Colorado State Univ., Fort Collins. 59 p.

Bokhari, U. G., J. S. Singh, and F. M. Smith. 1974. Influence of temperature regimes and water stress on the germinability of three range grasses and its possible ecological significance to a short-grass prairie. US/IBP Grassland Biome Preprint No. 101. Colorado State Univ., Fort Collins. 24 p. (Submitted to *Journal of Applied Ecology*.)

Brown, E. M. 1939. Some effects of temperature on the growth and chemical composition of certain pasture grasses. Missouri Agr. Exp. Sta. Res. Bull. 299. 61 p.

Brown, R. H., and R. E. Blaser. 1965. Relationships between reserve carbohydrate accumulation and growth rate in orchard grass and tall fescue. Crop Sci. 5:577-582.

Brown, R. H., and R. E. Blaser. 1970. Soil moisture and temperature effects on growth and soluble carbohydrates of orchard grass (*Dactylis glomerata*). Crop Sci. 10:213-216.

Bukey, F. S., and J. E. Weaver. 1939. Effects of frequent clipping on the underground food reserves of certain prairie grasses. Ecology 20:246-252.

Cook, C. W. 1966. Carbohydrate reserves in plants. Utah Res. Ser. No. 31. Utah State Univ., Logan. 47 p.

Davidson, J. L., and F. L. Milthorpe. 1966. Leaf growth in *Dactylis glomerata* following defoliation. Ann. Bot. (London) 30:173-184.

Ehara, K., N. Maeno, and Y. Yamada. 1966. Physiological and ecological studies on the regrowth of herbage plants. 4. The evidence of utilization of food reserves during the early stage of regrowth in bahiagrass (*Pospalum notatum* Flugge) with $C^{14}O_2$. J. Jap. Soc. Grassland Sci. 12:1-3 (from Herb. Abstr. 37:348).

Graber, L. F., N. T. Nelson, W. A. Luekel, and W. B. Albert. 1927. Organic food reserves in relation to the growth of alfalfa and other perennial herbaceous plants. Wisconsin Agr. Exp. Sta. Res. Bull. 80. 128 p.

Hutchinson, K. J. 1971. Productivity and energy flow in grazing/fodder conservation systems. Herb. Abstr. 41:1-10.

Hyder, D. N., and F. A. Sneva. 1959. Growth and carbohydrate trends in crested wheatgrass. J. Range Manage. 12:271-276.

Marshall, C., and G. R. Sagar. 1965. The influence of defoliation on the distribution of assimilates in *Lolium multiflorum* Lam. Ann. Bot. (London) 29:365-372.

May, L. H. 1960. The utilization of carbohydrate reserves in pasture plants after defoliation. Herb. Abstr. 30:239-245.

McIlroy, R. J. 1967. Carbohydrates of grassland herbage. Herb. Abstr. 37:79-87.

Murata, Y. 1969. Physiological responses to nitrogen in plants, p. 235-259. *In* J. D. Eashin, F. A. Haskins, C. Y. Sullivan, and C. H. M. Van Bavel [ed.] Physiological aspects of crop yield. Amer. Soc. Agron., Madison, Wisc.

Pearce, R. B., G. Fissel, and G. E. Carlson. 1969. Carbon uptake and distribution before and after defoliation of alfalfa. Crop Sci. 9:756-759.

Prianishnikov, D. N. 1951. Nitrogen in the life of plants. [Trans. by S. A. Wilde.] Kramer Business Service, Inc., Madison, Wisc. 109 p.

Smith, D. 1969. Removing and analyzing total nonstructural carbohydrates from plant tissue. Res. Rep. 41. Res. Div., Univ. Wisconsin, Madison. 9 p.

Smith, L. H., and G. C. Marten. 1970. Foliar regrowth of alfalfa utilizing C^{14}-labeled carbohydrates stored in roots. Crop Sci. 10:146-150.

Troughton, A. 1957. Chemical composition, p. 49-68. *In* A. Troughton [ed.] The underground organs of herbage grasses. Commonwealth Bur. Pasture Field Crops Bull. 44.

Trlica, M. J., and C. W. Cook. 1972. Carbohydrate reserves of crested wheatgrass and Russian wild rye as influenced by development and defoliation. J. Range Manage. 25:430-435.

White, L. M. 1973. Carbohydrate reserves of grasses: A review. J. Range Manage. 26:13-18.

Appendix I. Dry weight per pot (39.27 cm^2) for blue grama.

Days When Harvested	Temperature Regimes											
	13/7°C				24/13°C				29.5/18°C			
	C[a]	F[a]	I[a]	FI[a]	C	F	I	FI	C	F	I	FI
Shoots												
0[b]	3.20	3.51	3.40	3.47	2.87	3.25	3.61	3.53	3.31	3.27	3.25	3.68
45	4.72	5.22	6.72	7.23	5.63	5.45	7.53	8.72	6.22	5.37	7.81	8.86
65	5.83	6.32	8.32	8.78	6.74	6.20	9.30	11.37	7.46	5.87	9.72	12.82
90	7.25	7.82	9.82	10.25	8.55	7.54	12.42	15.27	9.34	7.26	12.84	16.32
120	8.30	8.71	10.42	11.32	9.25	8.35	13.35	16.87	10.27	8.20	13.55	17.38
Crowns												
0[b]	2.32	2.15	2.35	2.45	2.41	2.32	2.46	2.62	2.63	2.55	2.58	2.71
45	2.65	2.55	2.82	3.18	2.78	2.58	3.25	3.78	2.87	2.72	3.67	4.16
65	2.87	2.78	3.37	3.73	3.18	2.85	3.92	4.37	3.36	3.12	4.85	5.25
90	3.12	3.10	3.75	4.21	3.47	3.32	4.45	5.32	3.85	3.48	5.23	5.87
120	3.42	3.47	4.23	5.21	3.85	3.77	5.23	6.25	4.10	3.86	5.87	6.73
Roots												
0[b]	1.34	1.27	1.46	1.53	1.43	1.32	1.37	1.48	1.54	1.46	1.23	1.36
45	1.67	1.75	1.87	1.85	1.82	1.87	2.35	2.47	2.13	1.83	2.67	2.85
65	1.92	1.87	2.18	2.27	2.35	2.16	2.75	2.89	2.63	2.37	3.16	3.27
90	2.25	2.10	2.78	3.13	2.85	2.75	3.17	3.51	2.87	2.37	3.48	3.83
120	2.67	2.58	3.17	3.65	3.20	3.10	3.88	4.35	3.17	3.07	4.25	4.67

[a] C = Control; F = Fertilized; I = Irrigated; FI = Fertilized plus Irrigated.

[b] 0 day refers to the time when treatments were given.

Appendix II. Percent of TAC for blue grama plants.

Days When Harvested	Temperature Regimes											
	13/7°C				24/13°C				29.5/18°C			
	C[a/]	F[a/]	I[a/]	FI[a/]	C	F	I	FI	C	F	I	FI
Shoots												
0[b/]	6.45	6.35	6.85	6.75	6.69	7.12	7.23	7.23	7.85	6.69	6.81	7.12
45	7.65	8.25	10.25	12.25	7.75	8.75	11.75	14.63	8.75	9.83	14.35	16.35
65	10.25	8.85	13.85	15.69	10.85	9.26	15.60	18.65	12.35	9.27	18.25	22.83
90	10.85	9.25	15.75	18.32	11.87	10.83	17.25	20.20	12.84	9.87	20.50	24.60
120	9.12	8.23	11.30	13.20	8.75	7.83	13.75	15.32	8.00	7.75	15.25	16.81
Crowns												
0[b/]	2.42	2.47	2.45	2.52	2.48	2.47	2.40	2.42	2.45	2.52	2.41	2.46
45	2.57	2.83	3.75	4.45	2.87	2.85	4.18	4.86	3.23	2.75	3.87	5.37
65	2.87	2.85	3.87	4.25	3.12	3.17	4.85	4.86	3.15	2.62	5.12	5.78
90	2.32	3.13	4.12	4.55	3.45	3.37	5.87	6.58	3.02	2.87	6.15	6.78
120	3.75	3.48	4.87	5.25	3.55	3.42	6.35	7.74	3.67	3.38	6.86	7.87
Roots												
0[b/]	1.53	1.58	1.52	1.65	1.50	1.50	1.52	1.65	1.46	1.48	1.63	1.55
45	1.75	1.87	2.45	2.67	1.87	1.87	2.87	2.85	2.15	2.13	2.85	2.84
65	2.15	2.20	2.60	2.87	2.65	2.32	3.25	3.75	2.87	2.28	3.85	3.87
90	2.95	2.87	3.20	4.55	2.25	2.43	3.85	4.17	2.75	2.27	4.35	4.65
120	2.87	2.85	2.85	3.25	2.75	2.58	4.15	4.05	2.87	2.43	4.76	4.23

[a/] C = Control; F = Fertilized; I = Irrigated; FI = Fertilized plus Irrigated.
[b/] 0 day refers to the time when treatments were given.

Appendix III. Effect of various temperatures, water stress, and fertilization on the distribution pattern of labile energy in blue grama plants (kcal/pot).

Days When Harvested	Temperature Regimes											
	13/7°C				24/13°C				29.5/18°C			
	C[a]	F[a]	I[a]	FI[a]	C	F	I	FI	C	F	I	FI
Shoots												
0[b]	0.776	0.838	0.875	0.880	0.721	0.870	0.981	0.959	0.976	0.822	0.839	0.985
45	1.357	1.619	2.589	3.330	1.640	1.793	3.333	4.796	2.046	1.984	4.209	5.446
65	2.246	2.102	4.332	5.179	2.749	2.158	5.455	7.973	3.464	2.045	6.669	11.597
90	2.957	2.719	5.696	7.060	3.565	3.070	8.055	11.597	4.509	2.694	9.897	15.095
120	2.846	2.695	4.427	5.618	3.043	2.458	6.901	9.717	3.089	2.389	7.769	10.985
Crowns												
0[b]	0.211	0.199	0.216	0.232	0.224	0.215	0.221	0.238	0.242	0.241	0.233	0.250
45	0.256	0.271	0.404	0.532	0.299	0.276	0.510	0.690	0.348	0.281	0.534	0.839
65	0.309	0.297	0.490	0.596	0.373	0.339	0.714	0.798	0.397	0.307	0.933	1.140
90	0.389	0.364	0.580	0.720	0.450	0.420	0.982	1.316	0.437	0.375	1.209	1.496
120	0.482	0.454	0.774	1.028	0.513	0.484	1.248	1.818	0.565	0.490	1.514	1.991
Roots												
0[b]	0.077	0.075	0.083	0.094	0.080	0.074	0.079	0.092	0.084	0.081	0.075	0.079
45	0.109	0.123	0.172	0.185	0.127	0.131	0.253	0.264	0.172	0.146	0.286	0.304
65	0.155	0.154	0.213	0.244	0.234	0.188	0.336	0.407	0.283	0.203	0.457	0.475
90	0.249	0.226	0.334	0.441	0.241	0.251	0.458	0.550	0.296	0.240	0.569	0.669
120	0.288	0.276	0.339	0.446	0.330	0.300	0.605	0.662	0.342	0.280	0.760	0.742

[a] C = Control; F = Fertilized; I = Irrigated; FI = Fertilized plus Irrigated.
[b] 0 day refers to the time when treatments were given.

Appendix IV. Effect of various temperatures, water stress, and fertilization on the distribution pattern of nonlabile energy in blue grama plants (kcal/pot).

Days When Harvested	Temperature Regimes											
	13/7°C				24/13°C				29.5/18°C			
	C[a]	F[a]	I[a]	FI[a]	C	F	I	FI	C	F	I	FI
Shoots												
0[b]	12.984	14.252	13.744	14.039	11.618	13.100	14.539	14.210	13.253	13.237	13.130	14.835
45	18.932	20.821	26.300	27.750	22.559	21.637	29.036	32.693	24.694	21.105	29.370	32.643
65	22.813	25.067	31.437	32.570	26.230	24.501	34.535	40.917	28.606	23.194	35.120	44.116
90	28.212	30.900	36.523	37.009	33.224	29.350	45.344	54.062	35.651	28.516	45.313	55.075
120	32.844	34.755	40.373	43.052	36.727	33.442	50.498	62.822	41.071	32.871	50.490	63.745
Crowns												
0[b]	9.759	9.040	9.884	10.298	10.135	9.755	10.348	11.022	11.058	10.718	10.856	11.399
45	11.134	10.689	11.935	13.138	11.650	10.814	13.459	15.559	11.991	11.409	15.246	17.040
65	12.030	11.652	14.000	15.434	13.297	11.910	16.135	17.991	14.042	13.103	19.916	21.429
90	13.021	12.965	15.539	17.380	14.470	13.849	18.148	21.544	16.113	14.589	21.271	23.744
120	14.218	14.466	17.405	21.372	16.036	15.725	21.231	25.051	17.064	16.099	23.726	26.939
Roots												
0[b]	5.683	5.385	6.187	6.475	6.059	5.596	5.810	6.267	6.535	6.189	5.205	5.761
45	7.070	7.397	7.868	7.764	7.692	7.909	9.846	10.355	8.978	7.713	11.194	11.946
65	8.095	7.885	9.157	12.175	9.866	9.092	11.484	12.013	11.016	9.987	13.123	13.584
90	9.420	8.803	11.616	13.009	12.009	11.569	13.171	14.540	12.043	11.879	14.391	15.820
120	11.192	10.814	13.290	15.244	13.429	13.029	16.075	18.038	13.288	12.919	17.509	19.337

[a] C = Control; F = Fertilized; I = Irrigated; FI = Fertilized plus irrigated.
[b] 0 day refers to the time when treatments were given.