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# SOLUBLE CARBOHYDRATES, CONCURRENT PHOTOSYNTHESIS AND EFFICIENCY IN REGROWTH FOLLOWING DEFOLIATION: A FIELD STUDY WITH *AGROPYRON* SPECIES

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## SUMMARY

(1) The contribution of stored carbon to plant regrowth following defoliation is commonly thought to be large. This study quantitatively compared the amount of carbon supplied to regrowth from storage with photosynthesis of regrowth in the field.

(2) Two bunchgrass species were selected for comparison because of markedly different abilities to regrow following defoliation despite very similar photosynthetic, morphologic and phenologic characteristics.

(3) The more grazing-tolerant *Agropyron desertorum* (Fisch. ex Link) Schult. consistently produced more regrowth in the absence of photosynthesis than *A. spicatum* (Pursch) Scribn. & Smith, but a severe preclipping treatment (which has been shown to reduce carbohydrate reserves by more than 40%) did not significantly reduce etiolated regrowth in either species.

(4) Differences in regrowth between and within species were not correlated with crown non-structural carbohydrate concentrations, total pools, or amounts utilized during regrowth.

(5) The daily contribution of carbon from reserves exceeded measured daily photosynthetically-fixed carbon for only 2·5 days following defoliation when regrowth rate was maximal. However, when apical meristems were removed and regrowth was much slower, photosynthesis during regrowth immediately outweighed stored reserves as a source of carbon. The latter is the usual case when these grasses are subjected to managed grazing.

(6) In both species, apical meristem removal was followed by sharply reduced regrowth efficiency due to a delay in and reduced rate of regrowth production. There was, however, no increase in respiration rate.

(7) Meristematic limitations appear to be the dominant control on the amount of etiolated regrowth produced. These limitations also appear to be of prime importance in determining regrowth in the light and for grazing tolerance of plants.

## INTRODUCTION

Soluble carbohydrate reserves are often considered the primary source of carbon for regrowth following defoliation (Cook 1966; White 1973; Trlica 1977; Trlica & Singh 1979; Deregibus, Trlica & Jameson 1982). However, in several species there is little, if any, mobilization of reserves from roots to shoots following defoliation (Marshall & Sagar 1965; Davidson & Milthorpe 1966) and correlations between soluble carbohydrates and regrowth have frequently been unsuccessful (May 1960; Ward & Blaser 1961; Jameson 1963; Stoddart, Smith & Box 1975; Caldwell *et al.* 1981).

Possible causes for the lack of correlation between carbohydrate pools or concentrations and regrowth are: (i) the contribution of concurrent photosynthesis to regrowth is large; (ii)

morphological or meristematic features limit regrowth; or (iii) carbohydrate pools are inadequately assessed by common procedures. The importance of each of these factors was examined in field experiments utilizing two species that were selected because they differ greatly in tolerance of herbivory. These species, *Agropyron desertorum* (Fisch. ex Link) Schult. and *Agropyron spicatum* (Pursh) Scribn. & Smith,* have similar photosynthetic characteristics, the same growth form, and nearly identical phenology (Caldwell *et al.* 1981, 1983).

In addition to the confounding effects of concurrent photosynthesis and meristematic limitations on the relationship between regrowth and stored carbohydrates, there may be inherent differences in the efficiency with which the plant utilizes carbon for shoot regrowth. Following severe defoliation, *A. desertorum* allocates relatively more carbon to the shoot system and curtails root growth more than does *A. spicatum* (Caldwell *et al.* 1981; Richards 1984). *Agropyron desertorum* thus should have higher efficiency of carbon utilization for regrowth.

To examine the relationship between regrowth and soluble carbohydrates without confoundment by current photosynthesis, it was necessary to prevent photosynthesis since regrowing *A. desertorum* plants, which rapidly produce a large canopy, can fix up to twice as much carbon on a daily basis as regrowing *A. spicatum* plants (Caldwell *et al.* 1981). Thus, the etiolated regrowth technique (e.g. McKendrick & Sharp 1970; Christiansen, Ruelke & Lynch 1981) was used to provide a measure of the ability of the two grasses to mobilize stored reserves and synthesize new above-ground tissues.

Net $CO_2$ exchange measurements of severely defoliated bunchgrasses were conducted under field conditions to determine if the above-ground portion of severely defoliated plants could maintain a positive carbon balance, and to assess the potential contribution of photosynthesis to regrowth immediately following defoliation. Comparison of results of experiments in which active meristems were or were not removed allowed evaluation of morphological or meristematic limitations to regrowth.

## MATERIALS AND METHODS

Plants of the widely utilized Eurasian *A. desertorum* and the North American *A. spicatum* had been established in 1978 in experimental plots 4 km north-east of Logan, Utah (41°45'N, 111°48'W, 1460 m a.s.l.) in an area formerly dominated by *A. spicatum* and *Artemisia tridentata* ssp. *vaseyana* (Rydb.) Beetle. The *Agropyron* species are commonly defoliated heavily by large grazing mammals in semi-arid rangelands. Since grass response to defoliation is strongly influenced by competitors (Mueggler 1972), both species of grass were planted alternately within a uniform matrix of the *Artemisia* which provided a natural competitive background. Further site details are provided by Caldwell *et al.* (1981).

### *Etiolated regrowth experiments*

Fifty plants of each grass species were used for the etiolated regrowth experiments. Plants were randomly assigned to control or preclipping treatments and to date of initiation of etiolated regrowth. The preclipping treatment removed approximately 85% of the photosynthetic surface (5–7 cm stubble) on 30 April and 14 May 1981. This treatment caused a large reduction in the amount of stored carbohydrate. Crown carbohydrate

* Recent taxonomic revisions make *A. spicatum* synonymous with *Pseudoroegneria spicata* (Pursh) Löve (Löve 1980; Dewey 1984).

concentrations and pools were reduced 40–50% in *A. desertorum* and 55–65% in *A. spicatum* for the entire growing season by a similar severe defoliation treatment in 1980 (Caldwell *et al.* 1981) and comparable reductions in stored cabohydrate following defoliation have been reported by many other authors (e.g. Hanson & Stoddart 1940; Blaisdell & Pechanec 1949; Cook, Stoddart & Kinsinger 1958; Menke & Trlica 1983). Etiolated regrowth was initiated for five control plants of each species on 16 March and 16 April, and for five control and five preclipped plants of each species on 27 May, 18 June, 30 July and 27 October 1981. Etiolated regrowth was initiated by severely defoliating the plant and covering the tussock with an opaque plastic container.

Because both *Agropyron* species exhibit rapid internode elongation in early May, active apical and intercalary meristems were elevated and were removed in the defoliation at the initiation of the May, June and July etiolated regrowth experiments. When these active meristems were removed regrowth could only proceed after activation of basal (axillary) meristems. In the March, April and October experiments active meristems were not elevated and remained on the plants. Thus, these two groups of experiments represent different meristematic potential for regrowth.

The covers used in the etiolated regrowth experiments were painted white to prevent overheating. Crown temperatures of covered plants were not significantly different than those of uncovered plants in March–July or October, when most regrowth occurred. Soil water potential was at or above −0·1 MPa in the lower rooting zone of these plants until early July, and October rains recharged the upper profile. Thus, no significant water or heat stress occurred during periods when regrowth was proceeding in the dark.

Etiolated foliage produced in the dark was removed at frequent intervals, oven dried (80 °C, 24 h) and weighed. When regrowth ceased the plant was excavated, the crowns were washed free of soil, freeze-dried and weighed. Regrown foliage and crowns were each ground in a Wiley mill to pass a 40-mesh screen. Subsamples (50 mg) were then used for crown total non-structural carbohydrate (TNC) analyses and crown and foliage carbon content determinations.

At 2–3-week intervals during the 1981 growing season, other plants similar to the etiolated regrowth control plants were harvested to provide estimates of crown biomass, TNC concentration and pool size, and crude protein concentration and pool size at the beginning of each dark regrowth experiment. Shoots and crowns were freeze-dried, weighed and ground for analysis of TNC and Kjeldahl nitrogen. Crowns were also analysed for carbon content.

Although the diffuse root systems of both *Agropyron* species contain carbohydrates, there is no indication from studies with these and other grasses that root carbohydrates are mobilized for shoot growth (Marshall & Sagar 1965; Davidson & Milthorpe 1966; White 1973; Caldwell *et al.* 1981). In this study, carbohydrates available for shoot growth were considered to reside in the crowns, which included the lowest 1 cm of stem base and 1 cm of attached roots.

### *Carbohydrate and total carbon analyses*

Numerous extraction techniques have been used for determination of TNC. Each provides a different estimate of the amount of carbohydrate that can be mobilized easily for metabolism or translocation (Smith 1981). Two standard extraction techniques were used in this study; one (boiling water extraction) provided a conservative estimate of soluble carbohydrates and the other (boiling 0·2 N $H_2SO_4$ extraction) an overestimate of TNC (Smith 1981). Starch in the residue was digested using amyloglucosidase (Sigma

Chemical Co.) in the enzyme digestion technique of Haissig & Dickson (1979). Sugar concentration in all fractions was determined by the phenol-sulphuric acid method (Dubois *et al.* 1956). In these grasses the majority of TNC is fructosan (Smith 1968). Thus, fructose was used as standard. Starch concentrations were consistently very low (<0·5%) and were often not significantly different from zero. Thus, TNC concentrations (dry weight basis) given here include only the carbohydrate in the water or acid extract. TNC pools were calculated by multiplying plant part biomass by TNC concentration.

The carbon content of crowns and regrown foliage was determined by combustion in oxygen at 900 °C in the furnace of a carbon train (Lingberg, Sola Basic Industries). Water vapour was removed from the furnace exhaust by passing it through a column of Drierite (W. A. Hammond Drierite Co.). Carbon dioxide in the exhaust was then determined by absorption in a preweighed column of Ascarite (Arthur Thomas Co.). Carbon content, rather than biomass, was the basic unit for comparison of regrowth, respiration and photosynthesis.

### *$CO_2$ efflux during etiolated regrowth*

Relative $CO_2$ efflux rates during regrowth in the dark were determined on ten control plants of each species from May through July 1982. Carbon dioxide evolved beneath aluminium foil-covered metal containers was trapped by 40 g of soda lime following the procedure described by Minderman & Vulto (1973) and etiolated regrowth produced by these plants was harvested at frequent intervals, dried and weighed. The metal containers were pressed into the soil 2·5 cm and sealed around the perimeter with a saturated soil paste. The rate of $CO_2$ evolution was corrected by subtracting the amount of $CO_2$ evolved under nearby identical containers over bare soil. This soil component averaged 27% of the total $CO_2$ efflux. The daily plant $CO_2$ efflux rates determined by soda line trapping were of the same magnitude as rates determined by gas exchange measurements (see below) in the dark. Although the magnitudes of $CO_2$ efflux determined by soda line trapping were reasonable, only comparative $CO_2$ efflux values are reported, as suggested by Minderman & Vulto (1973).

### *Daily net assimilation following defoliation*

Carbon dioxide exchange of whole bunchgrass plants following severe defoliation was measured with a large cuvette system programmed to track ambient conditions of temperature and vapour pressure deficit as plants were exposed to normal solar radiation in May 1982. The heat exchanger and electronic control system were modified from an original Sirigor (Siemens Co.) chamber (Koch, Lange & Schulze 1971). Further details of this system are contained in Caldwell *et al.* (1981). Plants used for gas exchange measurements were severely defoliated by removing all foliage above 8-cm height and removing all remaining leaf blades. Thus, the only remaining photosynthetic tissue was leaf sheath material on the stubble. The plants were then subjected to continuous gas exchange measurement for 4 days.

## RESULTS

### *Regrowth produced from stored reserves*

Contrary to conventional expectation, the total amount of etiolated regrowth produced was not significantly affected by the severe preclipping treatment ($F < 0·93$, $P > 0·41$) (Fig. 1, inset). Because of this, further analyses were performed on the data for control

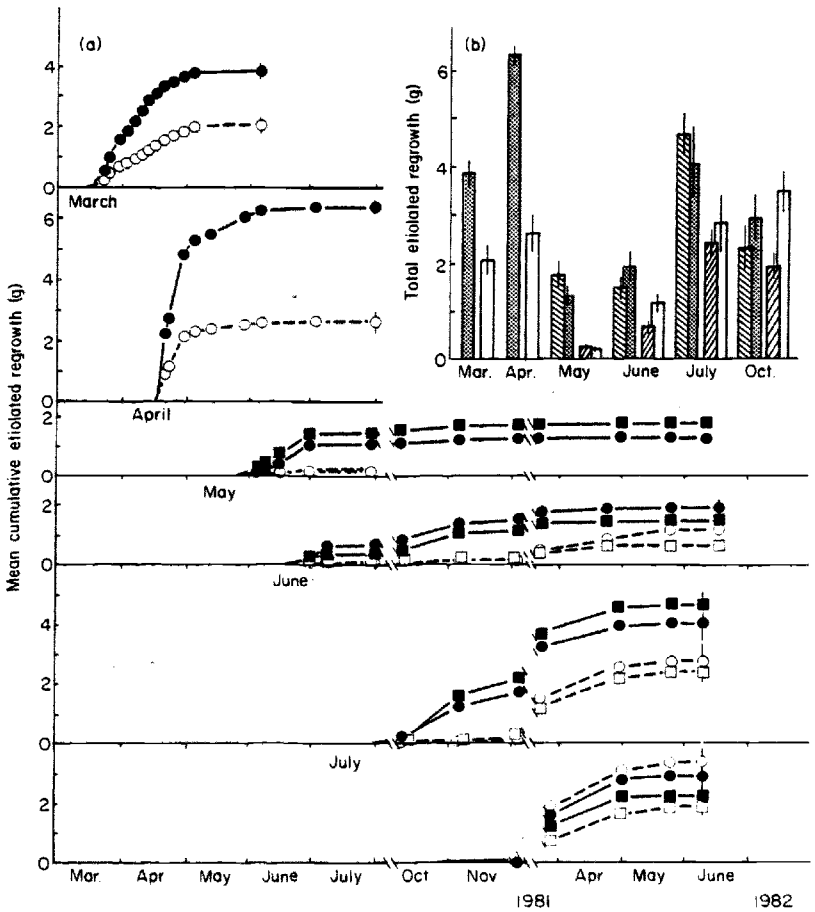


FIG. 1. (a) Time course of etiolated regrowth production in each of the six field experiments conducted in 1981 for control (●, ○) and preclipped (■, □) *Agropyron desertorum* (●, ■) and *A. spicatum* (○, □) plants. (b) Seasonal comparison of total etiolated regrowth (inset) for control (▩, □) and preclipped (■, ■) *A. desertorum* (▩, ■) and *A. spicatum* (□, ■) plants. Each point or bar represents the mean cumulative or mean total etiolated regrowth of five plants and ±S.E. is shown for the totals.

plants alone or as an unbalanced design, by combining the data for the preclipped and control plants for each species and date. These analyses gave similar results (Table 1).

The more grazing tolerant *A. desertorum* consistently produced more etiolated regrowth than *A. spicatum* except in October (Fig. 1, inset; Table 1). Etiolated regrowth production was lowest in both species in May and June and higher both earlier and later. In early spring, March and April, initial regrowth occurred rapidly in both species, but in the May and June experiments initial regrowth was much slower (Fig. 1). In these two experiments the rate of regrowth of *A. desertorum* increased after 2–3 weeks, whereas *A. spicatum* continued to regrow slowly. Coincident with the increased growth rate of *A. desertorum*, we observed the appearance of daughter tillers which arose from basal axillary buds. In other studies where tiller numbers were intensively monitored following defoliation, *A. desertorum* always produced daughter tillers more rapidly and in greater numbers than *A.*

*spicatum* (e.g. 0·71 and 0·04 daughter tillers per defoliated tiller, respectively, 2 weeks following a May defoliation (Caldwell *et al.* 1981)).

When etiolated regrowth was initiated in May or June, some regrowth occurred in the following fall and spring growing periods, and in the July and October experiments the majority of regrowth was produced in the following spring (1982) growing period (Fig. 1). This spring etiolated regrowth exhibited a time course similar to that seen in the March and April (1981) experiments. The normal seasonal growth pattern for both of these species is characterized by production of small tillers in the fall followed by rapid spring growth of those tillers. Regrowth produced in each season was correlated with the initial rate of regrowth in that season (Fig. 1).

### *Reserve availability and utilization*

Crown biomass and soluble carbohydrate availability at the beginning of each dark regrowth experiment were estimated by harvesting and analysing plants which were similar in size and age to the dark regrowth control plants. The harvested plants were growing in adjacent field plots of identical density, treatment history and with the same competitive environment of *Artemisia*. In both species crown biomass increased significantly through the growing season and declined during the summer drought period from July to October (Table 1). Crown carbohydrate pools followed a similar pattern; however, when weak acid was used as the extractant the pools appeared 2–3 times larger than when boiling water was used as the extractant. There were no significant differences between species in crown biomass or carbohydrate pools regardless of extraction technique used (Table 1). Crown carbohydrate concentrations exhibited seasonal patterns not dissimilar to the patterns exhibited by crown biomass and carbohydrate pools (Table 1). Concentrations, however, were significantly higher in *A. desertorum* than in *A. spicatum*.

As expected, crown biomass and crown carbohydrate pools were severely reduced during all dark regrowth experiments (Table 1). After regrowth ceased *A. spicatum* generally had smaller remaining carbohydrate pools, by both extraction methods, than did *A. desertorum*, but species comparisons within dates were statistically significant ($P < 0·05$) in only two of twelve comparisons (i.e., water-extracted pools in April and June).

Initial, water-extracted carbohydrate pools (and the change in these pools during regrowth) were often not large enough to account for the amount of regrowth produced. For example, in the March experiment, 3·81 and 2·06 g of regrowth were produced in the dark by *A. desertorum* and *A. spicatum*, respectively, but initial water-soluble carbohydrate pools were only 1·70 and 1·46 g, respectively (Table 1). However, the initial, acid-extracted carbohydrate pools (5·62 and 5·14 g, respectively) were large enough to account for the etiolated regrowth produced in all but one case (*A. desertorum*, April). Yet, compounds other than acid-extracted carbohydrates may have been mobilized because the change in crown biomass, which averaged 45% of the original biomass, was generally greater than the initial, acid-extracted carbohydrate pool, which averaged 35% of original crown biomass.

Nitrogenous compounds may have been one important constituent of the net carbon loss from the crowns that were not included in acid-extracted carbohydrates. The carbon content of the crude protein in the crowns was similar in both species and was equivalent to 11–15% of the carbon in acid-extracted carbohydrates in July and 26–34% in March (crude protein was calculated as 6·25 times Kjeldahl nitrogen and assumed to contain 53% carbon—Maynard & Loosli 1969).

TABLE 1. Crown carbohydrate and biomass parameters and shoot regrowth production† for control plants of *Agropyron desertorum* (Agde) and *Agropyron spicatum* (Agsp) in six 1981 etiolated regrowth experiments. Within columns, numbers followed by the same letter are not significantly different at $P = 0.05$ by Duncan's multiple range test.

| | | Initial conditions | | | | | Conditions after cessation of regrowth | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | | Crown soluble carbohydrates | | | | | Crown soluble carbohydrates | | | | Change in soluble carbohydrate pool | | |
| | | | Water extract | | Acid extract | | | Water extract | | Acid extract | | | | |
| Date of regrowth initiation 1981 | | Crown biomass (g) | Conc. (mg g$^{-1}$) | Pool (g) | Conc. (mg g$^{-1}$) | Pool (g) | Crown biomass (g) | Conc. (mg g$^{-1}$) | Pool (g) | Conc. (mg g$^{-1}$) | Pool (g) | Water extract (g) | Acid extract (g) | Change in crown biomass (g) | Shoot regrowth produced‡ (g) |
| 16 March | Agde | 14·7a | 114·5abc | 1·70a | 381·3bcde | 5·62ab | 8·0ab | 41·6cde | 0·34abc | 227·4ab | 1·83ab | 1·35 | 3·79 | 6·7 | 3·81de |
| | Agsp | 14·6a | 100·4ab | 1·46a | 353·0abcd | 5·14a | 6·0a | 41·9cde | 0·24ab | 251·8abcd | 1·51a | 1·22 | 3·63 | 8.6 | 2·06bc |
| 16 April | Agde | 16·8a | 114·5abc | 1·93ab | 314·6abc | 5·27a | 12·5abc | 55·7e | 0·69e | 271·1bcd | 3·39abcd | 1·25 | 1·88 | 4·3 | 6·33f |
| | Agsp | 25·6bcd | 81·6a | 2·09ab | 316·4abc | 8·11abc | 9·0ab | 46·6de | 0·42abcd | 196·4ab | 1·78a | 1·67 | 6·33 | 16·6 | 2·61cd |
| 27 May | Agde | 25·5bcd | 122·2bc | 3·13b | 336·8abc | 8·59bc | 13·2bc | 45·3cde | 0·61cde | 243·5abc | 3·21abcd | 2·52 | 5·38 | 12·3 | 1·29ab |
| | Agsp | 22·2abc | 95·4ab | 2·12ab | 285·7a | 6·33ab | 9·2ab | 58·6e | 0·53cde | 254·1bcd | 2·33abc | 1·59 | 4·00 | 13·0 | 0·15a |
| 18 June | Agde | 25·8bcd | 176·2e | 4·56c | 386·1cde | 9·96cd | 17·1c | 36·6bcd | 0·64de | 294·7cd | 5·04d | 3·92 | 4·93 | 8·7 | 1·92bc |
| | Agsp | 21·7abc | 155·8de | 3·39bc | 346·9abc | 7·53abc | 9·8ab | 20·4ab | 0·21a | 298·1cd | 2·92abcd | 3·18 | 4·61 | 11·9 | 1·18ab |
| 30 July | Agde | 32·7d | 139·1cd | 4·56c | 443·4e | 14·50e | 14·2bc | 17·8a | 0·27ab | 305·8d | 4·35cd | 4·29 | 10·15 | 18·5 | 4·06e |
| | Agsp | 28·9cd | 159·0de | 4·58c | 422·2de | 12·90de | 14·0bc | 18·8ab | 0·27ab | 260·2bcd | 3·63abcd | 4·32 | 8·56 | 14·9 | 2·81cde |
| 27 October | Agde | 18·5ab | 118·7bc | 2·20ab | 376·8bcde | 6·97abc | 14·9bc | 35·5abcd | 0·53bcde | 234·3ab | 3·50abcd | 1·67 | 3·47 | 3·6 | 2·93cde |
| | Agsp | 21·6abc | 115·5abc | 2·49ab | 302·9abc | 6·55ab | 17·4c | 28·1abc | 0·48abcd | 232·1ab | 4·03bcd | 2·01 | 2·52 | 4·2 | 3·46de |
| | | | | | | | | F values | | | | | | | |
| da | | 8·73*** | 12·54*** | 14·26*** | 8·03*** | 16·71*** | 4·36** | 11·06*** | 5·39*** | 4·90*** | 3·75** | | | | 21·56*** |
| sp | | 0·01 | 4·34* | 1·83 | 7·46** | 2·29 | 3·76 | 0·85 | 7·88** | 1·96 | 4·66* | | | | 31·61*** |
| da × sp | | 1·99 | 1·59 | 0·93 | 0·67 | 2·28 | 1·25 | 1·62 | 3·16* | 2·39 | 0·87 | | | | 5·70*** |

Significance: $*P < 0.05$, $**P < 0.01$, $***P < 0.001$ for data (da), species (sp), and date–species interaction (da × sp) effects.

† Soluble carbohydrates were extracted by boiling water or boiling 0·2 N $H_2SO_4$ and are expressed as concentrations (mg fructose equivalent per g dry weight of crown tissue) or pools (g fructose equivalent); crown biomass and shoot regrowth production are dry weights.

‡ Increasing sample size by combining control and preclipped plants of each species (see Results) caused all F values to increase and species means were significantly different at all dates except October.

*Carbohydrates, photosynthesis and regrowth*

*Efficiency of regrowth*

Carbon content of regrowth and of crown tissue (average for all dates: 37·5 and 37·7%, respectively) was determined for each date and used to calculate total carbon in regrowth biomass and change in carbon of crowns. One index of the efficiency with which a plant utilizes carbon for shoot regrowth is the ratio of carbon in regrowth to the loss of carbon from the crown of the plant during regrowth. This ratio was relativized to the highest value and is shown for each of the six 1981 experiments (Fig. 2). This carbon utilization efficiency was generally much greater for *A. desertorum* than for *A. spicatum*. In the three experiments, May, June and July, where all active apical and intercalary meristems were removed, *A. desertorum* had reduced carbon utilization efficiency, but such reduction was apparent for *A. spicatum* only in May and June. In the October experiment *A. spicatum* regrew as efficienctly as *A. desertorum*.

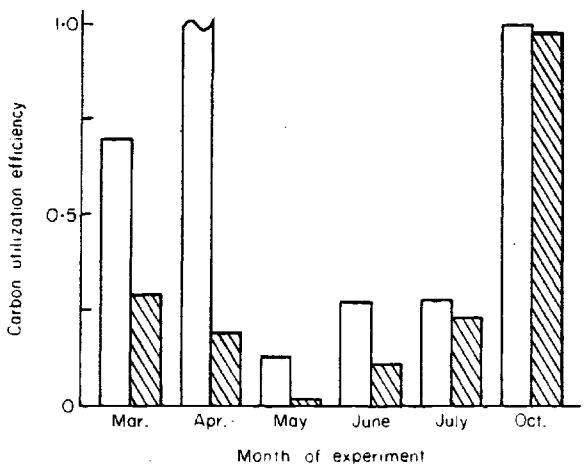


FIG. 2. Carbon utilization efficiency (carbon in regrown foliage divided by carbon lost from crowns) over the whole regrowth period, for experiments initiated in 1981 in the months indicated for *Agropyron desertorum* (□) and *A. spicatum* (■). In the April experiment *A. desertorum* regrowth carbon was greater than carbon lost from crowns (see Table 1), a result of necessarily using one set of plants to determine initial crown biomass and another set to determine regrowth and final crown biomass. Thus, carbon utilization efficiency, which was obviously high, was set = 1·0 in this case.

Another index of the efficiency of regrowth, shoot regrowth efficiency, is the ratio:

$$\frac{\text{regrowth C}}{\text{C efflux (as } CO_2\text{)} + \text{regrowth C}}$$

and is reported relative to the highest value. Carbon dioxide efflux was determined by 24-h sampling periods each week and interpolated for the total regrowth period (Fig. 3). When dark regrowth was initiated on 24 May 1982 active apical and intercalary meristems were not removed. After 1 week of regrowth these meristems were removed (Fig. 3) so that all further regrowth was from newly activated basal meristems. *Agropyron desertorum* had a higher $CO_2$ efflux rate than *A. spicatum* ($P < 0{\cdot}001$), but still was able to produce regrowth more efficiently. Removal of active meristems decreased $CO_2$ efflux rates of both species and caused a severe drop in shoot regrowth efficiency. Shoot regrowth efficiency

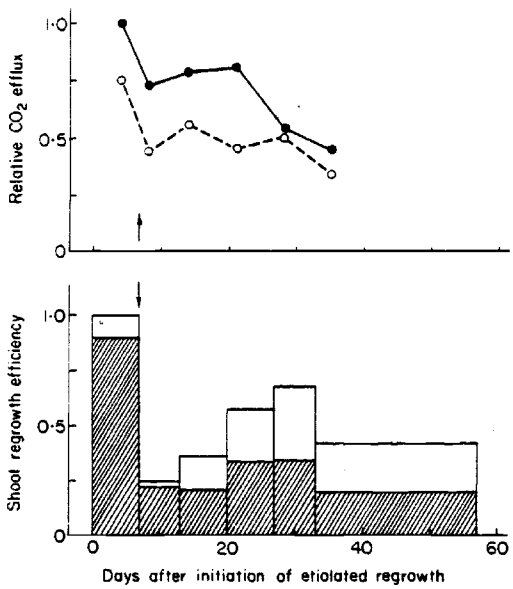


FIG. 3. Relative $CO_2$ efflux rates during single 24-h periods and calculated shoot regrowth efficiency (see text) for the same plants over approximately weekly intervals for *Agropyron desertorum* (●, □) and *A. spicatum* (○, ■). Etiolated regrowth was initiated on 24 May 1982 by removing all leaf blades and expanded leaf sheaths and covering the plants ($n = 10$). Arrows indicate the time of apical and active intercalary meristem removal.

recovered more rapidly in *A. desertorum* in parallel with greater production of new tillers from basal meristems. Thus, apical meristem removal caused reduced shoot regrowth efficiency not by increasing respiration losses but by causing a delay in the plant's ability to utilize available reserves for new regrowth. This result agrees with the seasonal pattern of carbon utilization efficiency shown in Fig. 2.

### *Photosynthesis following defoliation*

The maximum etiolated regrowth rate for both species occurred in the first few days of the experiment initiated in April (Fig. 1). This represents a situation where the carbon flux from stored reserves to shoot regrowth is maximal. In Fig. 4 this contribution from stored reserves is compared with the photosynthetic carbon gain of very severely defoliated plants during the first few days following a defoliation event. Daily net photosynthetic rates are based on field gas-exchange measurements of severely defoliated tussocks and are scaled to correspond with the average plant size of those in the etiolated regrowth experiment. The running mean of daily regrowth from the April etiolated regrowth experiment, expressed as carbon, plus the carbon utilized in nocturnal respiration of the above-ground plant parts (based on the gas-exchange measurements shown) is the maximum potential daily contribution of stored reserves to above-ground regrowth. This is represented in Fig. 4 by the hatched bars. They are, of course, conservative estimates of total stored carbon use since they do not include any growth or respiration of the root system. For perspective, the daily photosynthetic carbon gain and the carbon involved in daily growth of the above-ground portion of tussocks in the light are shown for the sixteenth day following severe defoliation (data from Caldwell *et al.* 1981).

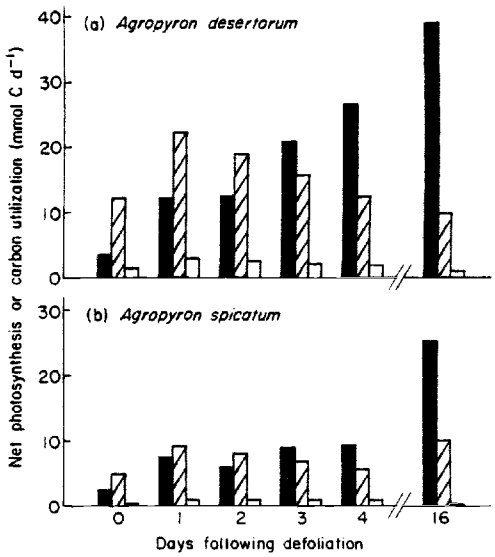


FIG. 4. Daytime net photosynthetic carbon uptake (■) of (a) *Agropyron desertorum* and (b) *A. spicatum* tussocks in the field following severe defoliation at midday on day 0, compared with daily carbon utilization (regrowth plus nocturnal respiration). (▨) represents stored carbon utilization when daily regrowth of severely defoliated tussocks in the field in the absence of photosynthesis (i.e. etiolated regrowth) was maximal (April experiments). (□) represents carbon utilization when etiolated regrowth rate paralleled regrowth rates more typically encountered following grazing. Daytime net photosynthesis and nocturnal respiration of the above-ground portion of defoliated plants is based on field $CO_2$-exchange measurements of entire tussocks. The gas exchange rates were scaled to correspond to plants of the same size as those used in the etiolated regrowth experiments.

If there are no meristematic limitations to regrowth and other conditions are conducive to rapid shoot growth, as in the April etiolated regrowth experiment, the potential contribution of carbon from reserves to shoot regrowth can exceed the photosynthetically fixed carbon of a very severely defoliated plant for a few days. This appears, however, to be an extreme case. When the carbon utilization for daily etiolated regrowth was plotted (Fig. 4) from experiments initiated at other times, a very different picture emerged. For example, in the experiment initiated in May, the carbon required for etiolated regrowth was less than 0·2 mmol carbon per day for both species, and the majority of carbon utilization was for nocturnal respiration. In this case, which is a more realistic simulation of the defoliation these species normally receive, total carbon utilized for regrowth plus dark shoot respiration was much less than the carbon provided by photosynthesis even on the day of defoliation for both species.

## DISCUSSION

### *Contribution of concurrent photosynthesis to regrowth*

While both *A. spicatum* and *A. desertorum* were able to produce substantial amounts of etiolated regrowth, up to 3 or 6 g per plant, respectively, these amounts represent only a small portion of the amount of regrowth produced in the light. For both species, regrowth in the light following clipping at various times through the growing season follows a seasonal pattern similar to the pattern found in this study for dark regrowth (Blaisdell &

Pechanec 1949; Cook, Stoddart & Kinsinger 1958) (Fig. 1, inset). When comparisons between dark regrowth and light regrowth following defoliation in different months (March–October) were made, dark regrowth ranged from 4 to 11% of light regrowth for *A. desertorum* and 1 to 9% of light regrowth for *A. spicatum*. Light regrowth amounts were obtained from severely defoliated plants used for another experiment at the same field site in 1981. If it is assumed that the amount of dark regrowth produced represents the maximum possible contribution of stored carbon to normal regrowth in the light, then it must be concluded that a least 89–99% of the carbon in regrown tissues of these species, under normal conditions, is derived from current photosynthate.

Under certain conditions, as following mid-April defoliation, the carbon contribution by stored reserves to shoot regrowth and respiration can exceed photosynthetic carbon gain of a tussock during the first few days following severe defoliation (Fig. 4). However, we consider this condition to be an unusual case. The plants used for gas exchange measurements were very severely defoliated as they possessed only sheath leaves on the remaining stubble, and no leaf blades. Furthermore, in this case, photosynthetic rates of these severely defoliated plants have been compared with short-term etiolated regrowth rates which were unusually high (Fig. 1). Therefore, under most field situations when less severe defoliation by grazing removes apical meristems, photosynthesis during regrowth would far outweigh stored reserves as a source of carbon, as also indicated in Fig. 4. It is commonly accepted that stored reserves are the major source of carbon for regrowth in these and other grasses (e.g. Hyder & Sneva 1959, 1963; Bokhari 1977; Daer & Willard 1981; Deregibus, Trlica & Jameson 1982). Our results contrast with this view, contribute to an explanation of why attempts to correlate soluble carbohydrates and regrowth have often been unsuccessful, and are in agreement with the laboratory studies of Marshall & Sagar (1965), Davidson & Milthorpe (1966) and the review of May (1960).

### *Soluble storage compounds*

Non-structural carbohydrates, which have been considered the most important carbon storage compounds (Cook 1966; White 1973; Deregibus, Trlica & Jameson 1982), are only a portion of the stored carbon which was utilized during dark regrowth in our experiments. The liberal estimates of change in soluble carbohydrate pool provided by the acid extraction procedure, which extracts some structural materials (Smith 1973), average only 52% (range: 31–96%) of changes in crown biomass (Table 1). Thus, compounds other than carbohydrates were probably utilized for regrowth, as has also been reported by Davidson & Milthorpe (1966), Chung & Trlica (1980), and Dewald & Sims (1981). The identity of these compounds remains unclear, but proteins, hemicelluloses, organic acids, etc., have been suggested. In this study, even if crude protein was completely utilized, it would not account for the discrepancy between the loss of carbon in acid-extracted carbohydrates and the loss of total carbon from the crowns. The participation of compounds apart from TNC as a source of regrowth carbon is apparent and could be one reason for previous difficulties in attempts to correlate TNC and regrowth (May 1960; Stoddart, Smith & Box 1975; Caldwell *et al.* 1981).

### *Relationship of soluble carbohydrates and etiolated regrowth*

Concentrations and pools of TNC found in *A. desertorum* and *A. spicatum* in this study are comparable to those reported in many previous studies of these and other perennial grasses in both absolute magnitude and in seasonal changes (Hanson & Stoddart 1940; Hyder & Sneva 1959, 1963; Daer & Willard 1981; Caldwell *et al.* 1981; Menke & Trlica

1981, 1983). Nevertheless, these values are poor indicators of etiolated regrowth production.

The amount of etiolated regrowth produced by *A. desertorum* and *A. spicatum* was not correlated with any measure of TNC pool or concentration in the plant at the beginning of the dark regrowth period or TNC or biomass lost during regrowth ($r < 0{\cdot}46$ and always N.S.) (Table 1). These two species are similar in amount and location of total carbohydrate pools; yet, the amount of etiolated regrowth produced was consistently greater in *A. desertorum* (Fig. 1, Table 1). This species also produces more foliage when regrowing in in the light than does *A. spicatum* (Hanson & Stoddart 1940; Cook, Stoddart & Kinsinger 1958; Caldwell *et al.* 1981). Furthermore, total carbohydrate pools and TNC concentrations were high when etiolated regrowth of these species was low (e.g. May and June, Table 1) and when TNC concentrations and pools were reduced due to preclipping, the production of etiolated regrowth was not reduced (Fig. 1).

### *Regrowth efficiency and meristematic limitations to regrowth*

Meristematic limitations to etiolated regrowth may provide the best explanation for the differences in regrowth between *A. desertorum* and *A. spicatum*. Nevertheless, even in the March and April experiments when regrowth was immediate and rapid from active apical and intercalary meristems in both species, more etiolated regrowth was produced by *A. desertorum*. This species utilized stored reserves more efficiently to produce new foliage or directed a larger proportion of the mobilized compounds to foliage-producing meristems (Fig. 2), perhaps at the expense of root growth (Richards 1984). The greater efficiency of *A. desertorum* was not due to lower $CO_2$ efflux rates of whole plants (Fig. 3).

Meristematic limitations, or limited flexibility for reallocation of resources (Watson & Casper 1984), appear to be much more important than the amount of stored or photosynthetically fixed carbon in determining the ability of these grasses to regrow following defoliation. Much past research has considered only one species or time of defoliation, or has compared species with different growth form or phenology. This may have obscured the importance of developmental constraints on allocation of carbon resources. Study focused on the characteristics which allow grazing-tolerant species, such as *A. desertorum*, to rapidly adjust carbon allocation in favour of shoot regrowth following apical meristem or canopy removal, is needed. This field study showed clearly that photosynthesis of regrowth was quantitatively the most important, and, under most conditions, nearly the sole source of carbon for shoot regrowth. The efficiency with which a species directs available carbon to above-ground growing points and utilizes it for synthesis of new foliage may be a key physiological feature which determines that species' ability to tolerate defoliation by large grazing animals.

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