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Diurnal Changes in Allocation of Newly Fixed Carbon in Exporting Sugar Beet Leaves[1]

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ABSTRACT

Storage of newly fixed carbon as starch and sucrose follows a regular daily pattern in exporting sugar beet leaves under constant day length and level of illumination. Up to the final two hours of the light period, when starch storage declines, a nearly constant proportion of newly fixed carbon was allocated to carbohydrate storage, principally starch. Sucrose is stored only early in the light period, when there is little accumulation of starch. Pulse labeling with $^{14}CO_2$ revealed that considerable starch synthesis was taking place at this time. Starch made the previous day was not mobilized during this period but breakdown of newly synthesized starch may occur when carbon flow into sucrose synthesis increases early in the day. At the end of the day, starch storage declined from the constant level observed during most of the day, but no diversion of label into export of specific alternative compounds could be detected. Lowered storage of starch persisted when the 14-hour light period was lengthened. Changed allocation of recently fixed carbon to sucrose and starch at the beginning and end of the light period was not the result of outright inactivation of pathways but of regulation of carbon flow.

Source leaves participate in control of export by allocating recently fixed carbon to a number of alternative uses (5). Partitioning of exported carbon among sinks has been found to differ between day and night (2, 7) and, to the extent that this occurs, distribution of newly fixed carbon between sucrose and starch affects partitioning among sinks. Consequently, regulation of carbon allocation in source leaves has an impact on translocation and, along with source and sink strength, is a potential determinant of crop yield.

In sugar beet, the proportion of newly fixed carbon allocated to sucrose and starch is adjusted at the beginning and end of the day while during most of the day distribution to these compounds is steady under a range of conditions (2). Study of these patterns of carbon allocation can help us understand adaptive priorities for carbon use during the diurnal cycle. Because these patterns of carbon use are the result of the particular control systems at work, they also can help us to understand mechanisms regulating translocation.

Recently, investigators have proposed a number of mechanisms for regulation of allocation to sucrose and starch synthesis (8, 13, 14). These regulatory processes are thought to coordinate sucrose synthesis rate with NCE[2] rate and to control storage of sucrose in exporting leaves. By moderating sucrose formation, these processes allow starch synthesis even in the presence of rapid synthesis and use of sucrose (13, 14). To relate mechanisms such as these to control of carbon allocation in exporting leaves, it is important to study responses of leaves in detail as they adjust to a variety of conditions, including the daily light-dark cycle.

In this work, we studied carbon allocation and various aspects of physiology in exporting sugar beet leaves, particularly at the beginning and end of the day. The data point to priority uses for recent products of photosynthesis and help characterize the regulatory mechanisms at work.

MATERIALS AND METHODS

Plant Material. Plants of a multigerm variety of *Beta vulgaris* L., Klein E were raised in a 14-h light period (25°C, 17°C, 450 μmol photons m^{-2} s^{-1} PAR) and used when 6 to 8 weeks old. Growth conditions and plant characteristics were similar to those described previously (2).

Pulse Labeling. Synthesis rates for starch and sucrose were estimated by pulse labeling recent products of photosynthesis with $^{14}CO_2$ for a short time to minimize turnover. In the first set of experiments, only the underside of the leaf was exposed to $^{14}CO_2$. To obtain a time course, cups were attached to several 1.7-cm^2 areas of mature leaves under a photon flux density of 425 μmol m^{-2} s^{-1}. A 0.1-ml mixture of 350 μl L^{-1} CO_2 and 74 kBq of $^{14}CO_2$ was injected into each cup which contained 5 ml of air stirred with a magnetic stir bar during the 2-min labeling period. After each pulse, a 0.78-cm^2 area of leaf was excised and treated as described below.

Immediate measurement of NCE of the leaves was impractical so determinations were carried out either the day before or after labeling. Because the top surface of the leaf was exposed to atmospheric CO_2 and the undersurface to $^{14}CO_2$, SR of the fixed gas was estimated from the NCE data and the ^{14}C fixed by the leaf area rather than from the gas supplied.

In later experiments, both surfaces of the leaf were exposed for 3 min to $^{14}CO_2$. Gas was circulated in a closed system through an IR gas analyzer and an ion chamber before it flowed over the top and bottom of the leaf area. Thus, SR of the gas and the amount of $^{14}CO_2$ fixed during each pulse were determined. The SR from each method and the radioactivity in the respective compounds were used to estimate the rate of synthesis of sucrose and starch from the recently fixed $^{14}CO_2$. The data resulting from pulse labeling allow us to estimate the rate of synthesis; steady-state labeling, by contrast, measures the rate of net accumulation. The pulse method of determining synthesis rates requires that the SR of the CO_2 closely approximates the SR of the ^{14}C entering the pool in question. The pools of the precursors of starch and sucrose likely are small and turning over rapidly, favoring existence of the assumed condition. If the SR of these precursor pools is lower than that of the CO_2 supplied, the synthesis rates would

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[2] Abbreviations: NCE, net carbon exchange; SR, specific radioactivity.

be underestimated. While not as quantitative as results from steady-state labeling, the estimates from pulse labeling are deemed reliable enough to indicate approximate rates of synthesis.

Steady-State Labeling. Depending on experimental design, one or two source leaves were enclosed in a chamber that was part of a closed system for supplying labeled CO_2. During a 14-h photoperiod, the leaf was supplied $^{14}CO_2$ maintained at approximately 380 $\mu l\ L^{-1}$ CO_2 with an average SR of 44.4 Bq g^{-1} C. The lid of the chamber was designed so that samples could be removed from the leaf with only a slight transient change in the SR of the gas. Accumulation of ^{14}C in various pools of the leaf was followed during the 14-h photoperiod by removing samples at 30- to 45-min intervals during the first and last 2 h of the light period and at intervals of up to 2 h during the intervening hours.

In another set of experiments with translocating and nontranslocating (heat-girdled) leaves, the photoperiod was extended by 4 or by 10 h. Heat-girdling a petiole near the blade was observed to stop export of ^{14}C from the blade completely as verified by a GM detector positioned next to the source leaf blade. Accumulation of ^{14}C in the source leaf was monitored while the plant photosynthesized in $^{14}CO_2$ for 8 h; there was no detectable loss of label from the blade when the supply of $^{14}CO_2$ was replaced with unlabeled CO_2. NCE, accumulation of ^{14}C in the source leaf, and arrival of label in a sink leaf were followed as described previously (2). Samples were taken from the leaves and analyzed as described below.

Mobilization of ^{14}C from starch synthesized during the light period was studied by allowing a leaf to photosynthesize in $^{14}CO_2$ under steady-state conditions for the 14-h day period. Samples were removed from the leaf 5 min before the end of the light period and at intervals during the last 2 hr of the dark period and the first 5 h of the next light period. The leaf was kept in room air during this second light period.

Extraction and Analysis of Carbohydrates. Excised leaf discs were immediately plunged into a mixture of 1:4 chloroform:methanol (v/v) at 80°C and then extracted in 80% ethanol. Extracts containing the soluble components of the leaf were analyzed for total sucrose by the nonamplified enzyme-based assay procedure of Outlaw and Manchester (10) as described earlier. Then the soluble extract was separated into ^{14}C-labeled lipid, neutral, organic acid, and amino acid components.

The extracted homogenized disc constituted the insoluble fraction which was analyzed for labeled and total starch. Radioactivity released with amyloglucosidase was considered to be ^{14}C-glucose from labeled starch. The amount of ^{14}C in the starch pool was determined by liquid scintillation counting of this fraction (1, 2).

Fractionation of Ethanol-Soluble Extract. Chloroform and water phases from chloroform:alcohol extract were separated by adding water. After the chloroform phase was removed, samples containing the water-soluble fraction were placed in heating blocks at 50°C, and evaporated to dryness in a stream of air. The residue, dissolved in water, was separated into neutral, acidic, and basic fractions as described by Redgwell (11). Columns containing 1 ml of Sephadex QAE-A-25 and Sephadex SP-C-25 (Pharmacia) were prepared with the SP column mounted in series above the QAE column. An aqueous, 0.5-ml sample from 0.65 cm^2 of leaf was layered on the SP Sephadex column and washed into it with 0.5 ml of H_2O. Neutral, acidic, containing organic acids, and basic, containing amino acids, fractions were eluted with 10 to 12 ml of H_2O, 4% (v/v) formic acid, and 0.2 M NH_4OH, respectively. Radioactivity in these fractions was determined by liquid scintillation counting. The neutral fraction was evaporated to dryness in a stream of air then redissolved in 0.3 ml of 80% ethanol. Components of the neutral fraction, including sucrose, were separated by TLC (3).

RESULTS

Generalized Pattern of NCE and Carbohydrate Accumulation. Data for NCE and for starch and sucrose accumulation were collected from eight experiments, some of which were described in an earlier publication (2). Data from the sets were averaged, yielding representative maximum values of 250 and 40 $\mu g\ C\ cm^{-2}$ for starch and sucrose content, respectively, and 1.50 $\mu g\ C\ cm^{-2}\ min^{-1}$ for the highest rate of NCE (Fig. 1). Smoothed curves of averaged NCE rates were integrated to show the total carbon fixed by mature sugar beet source leaves during a 14-h photoperiod.

Data in Figure 1 show the progress of NCE and storage of sucrose and starch throughout a 14-h light period in representative 6-week-old sugar beet source leaves. NCE rate reached a maximum during the first 2 h then declined slowly for the remainder of the light period. Recently fixed carbon accumulated rapidly and in a nearly linear fashion from the start of the day, but starch accumulation remained near zero for some time. The rate rose only gradually, becoming steady during the third hour of the light period. At the start of illumination, sucrose quickly increased above the level at night; by 2 h, accumulation had slowed to zero and remained there for the rest of the day. Cumulative NCE amounted to 1000 $\mu g\ C\ cm^{-2}$ after 14 h. Starch plus sucrose, totaling 290 $\mu g\ C\ cm^{-2}$, accounted for 29% of the fixed C. Export in these experiments averaged approximately 50% of the cumulative NCE (2). Approximately one-fifth of the fixed C was allocated to other assimilate pools in the leaf.

Allocation Early in the Day. During the first 2-h period, a gradual acceleration in starch accumulation generally paralleled the decline in sucrose storage (Fig. 1). If starch had accumulated at the rate of 30% of NCE observed later in the day, an additional 40 $\mu g\ C\ cm^{-2}$ leaf would have accumulated in this pool. The deficit was nearly equal to the 32 $\mu g\ C\ cm^{-2}$ initial increase in source leaf sucrose. Stored sucrose plus starch was a nearly constant proportion of both carbon fixed, approximately 25%, and of that part of recently fixed carbon not exported, about 40%. A major portion of the newly fixed carbon outside these two categories is exported.

The rate of entry of ^{14}C into sucrose and starch from a 2- to 3-min pulse of labeled CO_2 was used to estimate synthesis rates for these compounds during the initial phase of the light period. The data revealed similar synthesis rates for both sucrose and starch early in the light period (Fig. 2). Increase in sucrose synthesis corresponded to the increase in photosynthesis rate and with accumulation of additional sucrose in the leaf. Synthesis of

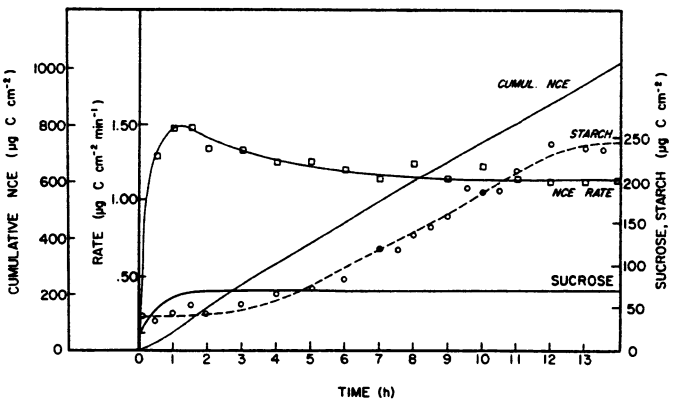

FIG. 1. Comparison of NCE rate, cumulative NCE, accumulation of sucrose, and storage of starch in 6-week-old sugar beet source leaves during a 14-h day. Data are averages from eight experiments of this type. Cumulative NCE was obtained by integration of values for NCE rate and is the total carbon fixed during the light period.

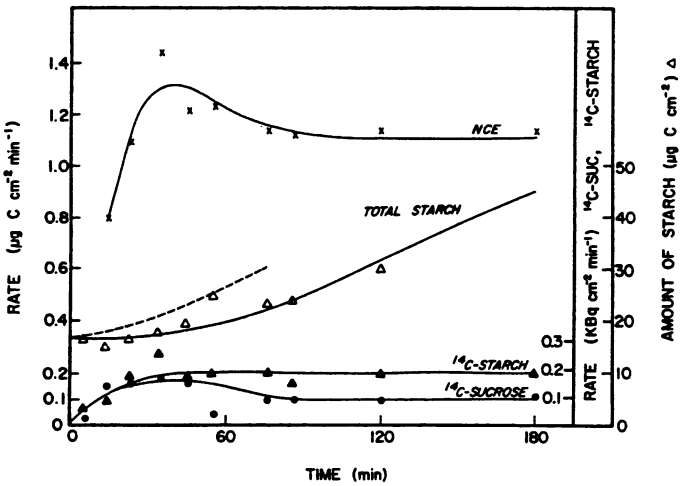

FIG. 2. Rates of NCE, ^{14}C-sucrose synthesis, ^{14}C-starch synthesis, and total amount of starch accumulated in the sugar beet source leaf during the first 3 h of the day. Each value for ^{14}C-starch and ^{14}C-sucrose synthesis rate was obtained by supplying a pulse of $^{14}CO_2$ to the undersurface of the leaf for 3 min. Rates are expressed as μg C cm^{-2} min^{-1} (left axis) and kBq cm^{-2} min^{-1} (right axis). Dashed line shows the amount of starch expected to be accumulated based on the rates of ^{14}C-starch synthesis. Data are representative values from one of two experiments of this type.

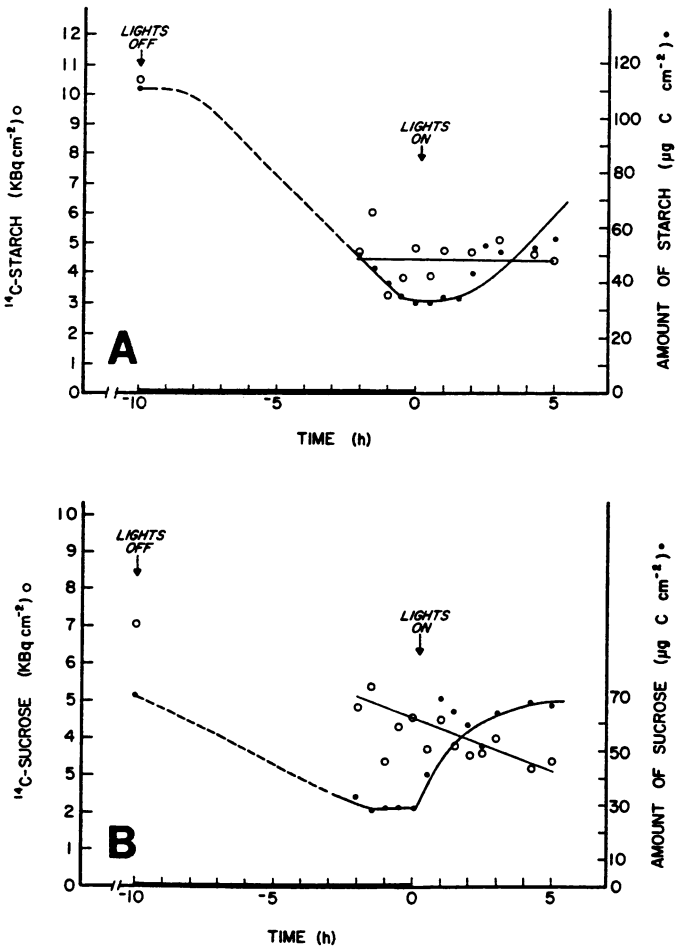

FIG. 3. Mobilization of ^{14}C from the starch (A) and sucrose (B) pools of a source leaf during the dark period and after the start of the light period. Time courses for depletion and accumulation of carbon in both pools and regression lines for ^{14}C also are shown. The leaf photosynthesized in $^{14}CO_2$ for 14 h before the first sample was taken then in room air during the dark and second light periods. Values are representative of data from two experiments of this type.

starch from newly fixed carbon began within minutes after lights were turned on. The value estimated by pulse labeling reached 60% of the midday rate within 20 min. This rate should have resulted in an increase in starch of 10 μg C cm^{-2} after 60 min in the light (dashed line, Fig. 2) but there was not a corresponding accumulation of starch initially. The increase actually took 120 min, suggesting that initially there was degradation of newly made starch.

Steady-state labeling was used to test the hypothesis that lessened starch accumulation resulted from starch turnover. Loss of ^{14}C from starch was followed at the end of the night and the beginning of the day in a leaf that had been supplied with $^{14}CO_2$ for 14 h the previous day. ^{14}C was lost from the starch pool during the following dark period but not after the initiation of the next light period (Fig. 3A). In contrast, ^{14}C was lost from sucrose throughout the period studied (Fig. 3B). This loss was expected for a translocating plant.

Allocation during the Day. Allocation of newly fixed carbon throughout the day was examined by steady-state labeling (Fig. 4). We studied accumulation of ^{14}C in the insoluble pool and the neutral, organic acid, amino acid, and lipid fractions of the soluble pool. Several pools such as organic acid and lipid, required several hours to attain isotopic saturation. In these cases, increased ^{14}C content reflected both the approach to isotopic equilibrium and net accumulation. In contrast to distribution of newly fixed carbon at the beginning and end of the day, during the main part of the day there was a relatively steady net flow of ^{14}C into starch and various pools of the soluble fraction in the leaf.

A definite lag in storage of labeled insoluble material (Fig. 4), similar to that observed for starch accumulation (Fig. 1), occurred at the start of the day. Concurrence between accumulation of total starch, determined chemically, and ^{14}C-starch, determined by dividing ^{14}C-starch by the SR of the CO_2 supplied, was also observed during the first 2 h of the light period. If labeling began 8 h after initiation of the light period, no lag in accumulation of ^{14}C in the insoluble fraction was observed. Initial rates of starch synthesis determined from these steady-state labeling data were 0.08 μg C cm^{-2} min^{-1} compared to 0.1 μg C cm^{-2} min^{-1} measured by a 3-min pulse of $^{14}CO_2$ supplied to both surfaces of the leaf.

Allocation Late in the Day. Data in Figure 1 indicate that NCE gradually decreased by approximately 25% following the maximum early in the day. By contrast, accumulation of starch remained steady during most of the day before it declined during the last 90 min. If allocation of carbon to the starch pool had continued at the midday rate, an additional 24μg C cm^{-2} would have accumulated in this pool by day's end. When the day was extended by 4 h, starch accumulation failed to increase from the end-of-day decline and the deficit reached 57 μg C cm^{-2} (Fig. 5). Raising the CO_2 concentration just enough to prevent the usual gradual decrease in NCE caused the rate of starch accumulation to remain steady right up to the end of the day. In this case, accumulation coincided in shape (but not exact amount) with the curve derived from cumulative NCE as shown in Figure 5.

In this study, we were not able to identify the fate of the carbon diverted from starch storage late in the day. There was no marked increase in label incorporated into the soluble fraction when starch accumulation slowed (Fig. 4). No major shifts in allocation of carbon to the neutral, organic acid, amino acid, or lipid pools were detected, either during the last 90 min of the day, or when the day was extended by 4 h.

Slowing of starch storage followed a pattern suggestive of a diurnal rhythm. Steady-state labeling studies showed that extend-

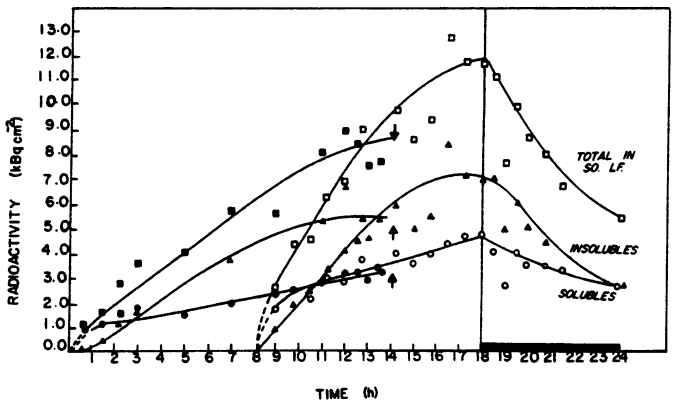

FIG. 4. Accumulation of ^{14}C in various pools of sugar beet source leaves given the usual 14-h photoperiod (closed symbols) and given a light period that was extended by 4 h (open symbols). Leaves photosynthesized in $^{14}CO_2$ maintained at a constant specific radioactivity before and after the brief sampling periods. Total in a source leaf is sum of ^{14}C accumulated in compounds of soluble plus insoluble fractions. Mobilization of ^{14}C from these pools is shown for the subsequent dark period in the plant given an extended light period. Arrows indicate usual beginning of the dark period. Each set of data is from one of two experiments of this type.

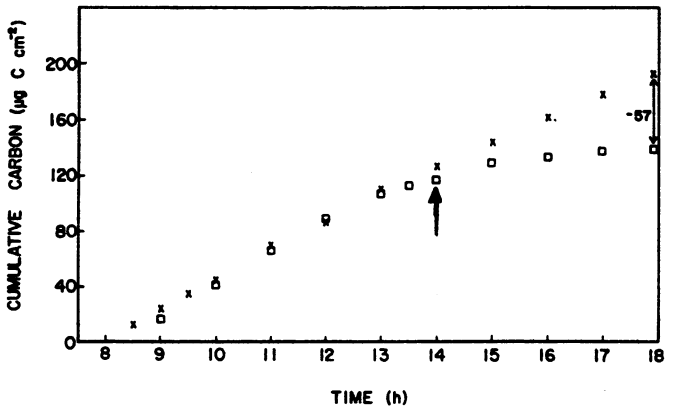

FIG. 5. Pattern of starch accumulation when the 14-h light period is extended by 4 h. Starch accumulated after the first sampling at 8 h was determined by subtracting the amount present at 8 h (94 μg C cm^{-2}) from each subsequent data point. Arrow indicates usual beginning of the dark period. The amount of starch (×) that would have accumulated if starch had continued to be stored at the midday rate of 30% of NCE was computed from cumulative NCE. Data are from one of two experiments of this type.

ing the light period a full 10 h had no effect on rates of NCE or accumulation of ^{14}C in the source leaf established near the end of the usual light period and, by inference, on export from the source leaf. Arrival of label in the monitored sink leaf was also steady during the decline in starch accumulation and on into the extended light period. Carbohydrate analysis of leaf samples revealed that starch accumulation did not resume in these plants (Fig. 6). We studied the effect of an extended light period in leaves girdled after 10 h of photosynthesis in $^{14}CO_2$. NCE remained constant after girdling. Both sucrose and starch began to accumulate after export was stopped (Fig. 6).

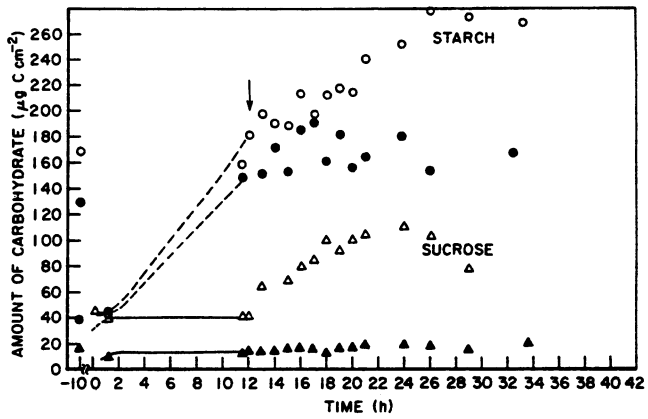

FIG. 6. Accumulation of starch and sucrose in sugar beet source leaves when the light period was extended to 24 h. One petiole on the plant was girdled after 12 h of light (open symbols) and the other was left intact (closed symbols). Data are from one of two experiments of this type.

DISCUSSION

Beginning of the Day. The fact that the rapid increase in sucrose early in the morning coincided with delayed starch accumulation (Fig. 1) points to a possible link between these events. A portion of newly fixed carbon, allocated to storage, may have been diverted to sucrose because of delayed activation of the starch pathway following the night. This explanation seems unlikely because, as revealed by early incorporation of newly fixed CO_2 into starch (Fig. 2), starch synthesis occurred in source leaves shortly after the start of the day. Although the rate was somewhat lower early in the day, starch synthesis was appreciable even when accumulation was nearly zero.

Storage of sucrose and starch likely constitute alternative uses of most of the newly fixed carbon not exported. This interpretation was borne out by the fact that the amount of sucrose accumulated was similar to the deficit in starch storage. Some carbon diverted from starch storage may have been used in other ways, such as export, but the sum of stored sucrose plus starch was rather uniform throughout the day. A nearly constant fraction was distributed to these two storage forms whether considered as the fraction of current NCE or of the unexported portion of the latter. Carbon allocated to storage appeared to be regulated to give a steady amount throughout the day even though distribution between the two forms changed.

At the start of the day, there appeared to be an initial priority for storage of a certain amount of sucrose and a resulting diversion of carbon from starch storage. This response may correspond to increased use to restore vacuolar sucrose depleted during the night (9). Rates of synthesis determined by pulse labeling confirmed that sucrose synthesis was more rapid at the time sucrose accumulated during the first hours of day (Fig. 2) and slowed concurrently with the gradual cessation of sucrose accumulation (Fig. 1). Results from the study of turnover (Fig. 3) show that starch stored the previous day was not mobilized to provide carbon for sucrose synthesis but turnover of newly synthesized starch could not be ruled out. Considerable incorporation of ^{14}C into starch by short-term labeling (Fig. 2) but not by steady-state labeling (Fig. 4) at the start of the day is consistent with turnover of the newly synthesized starch at that time.

As sucrose reached a critical level, ascendancy of control seemed to shift toward a steady rate of starch accumulation. Increased carbon use for sucrose synthesis may have caused breakdown of the most recently made starch resulting in a lag in storage. Synthesis of starch with little or no net accumulation (Fig. 2) lends support to simultaneous synthesis and degradation of recently made starch although this appears not to occur later in the day (3).

In some species such as barley, the concentration of sucrose in leaf cells may serve as a signal triggering synthesis and degradation of starch (6). While the gradual increase in starch accumulation noted early in the day supports this model, absence of a

decrease in sucrose accompanying the slowing of starch synthesis at the end of the day (Fig. 1) is not in line with this mechanism. Programmed, diurnal regulation of starch synthesis has a more direct influence on the allocation of carbon to starch than does competition for carbon destined for sucrose synthesis.

The size of the sucrose storage pool does seem to be controlled, in part, in relation to the state of carbon nutrition of the leaves (4, 9). There is evidence that a key enzyme of the sucrose synthesis pathway, sucrose phosphate synthase, is inhibited under some circumstances by cytoplasmic sucrose (12, 14) but the level of metabolites or effectors other than sucrose in the mesophyll cytoplasm also seems to regulate sucrose synthesis (13, 14).

End of the Day. The final hours of the day were characterized by a gradual relaxation of the control that maintained a constant rate of starch accumulation during most of the day. Decline of starch storage to near zero during the final 2 h (Fig. 1) was not a result of competing sucrose accumulation because sucrose level remained the same or, in some cases, increased only slightly. There was no evidence of increased export. The observation that starch mobilization only begins 60 to 90 min after the start of darkness (2) suggests that flow of carbon through the degradative pathway did not occur until after darkness and was not responsible for the decline in starch accumulation in the light. Experiments in which a leaf photosynthesized in $^{14}CO_2$ for 8 h then in $^{12}CO_2$ for the final 2 h of the light period showed that ^{14}C is not lost from the starch pool during the chase period (1). These observations make turnover of newly synthesized starch at the end of the day seem unlikely.

Nor was there extensive inactivation of the starch synthesis pathway in anticipation of the dark period. It is possible that decreased accumulation resulted from a lower rate of starch synthesis, an interpretation supported by data from pulse labeling. Leaves were found to be capable of starch synthesis but at a rate lower than that in the main part of the day. The reduction in starch synthesis may be seen as adaptive in view of the approaching period of starch utilization. Diversion of carbon to the amino acid pool in anticipation of increased protein synthesis during the night, for instance, seems logical. Distribution of labeled carbon during this period failed to reveal any obvious alternative destination for the carbon not accumulated as starch, neither the soluble fraction nor any specific major component (Figs. 1 and 5). The carbon probably was distributed among a number of products, making disposition of the diverted carbon hard to trace. Translocation, as evidenced by both export and import, did not show a change during the transition and on into the extended light period.

During much of the day, regulation of starch accumulation appeared to be oriented toward steady storage of starch rather than to allocation of a fixed proportion of NCE to starch. The proportion stored as starch gradually increased during the main part of the day, going from 0.30 several h after the start of the day to 0.32 approximately 2 h before its end. At that point, the value decreased steadily to near 0. Apparently as a result of an endogenous, diurnal rhythm, steady storage of starch gave way to decreased accumulation 1 to 2 h before the usual start of the dark period and was not reversed when the light period was extended (Fig. 4). The decrease was not inevitable; raising CO_2 concentration just enough to prevent the usual decrease in NCE caused the rate of starch accumulation to remain steady right to the end of the 14-h day. Reduced storage also was reversed when export of newly fixed carbon was stopped by petiole girdling (Fig. 6).

Control Mechanisms. Sucrose level or aspects of metabolism related to its synthesis have been proposed as key to the control of sucrose synthesis (5, 8, 13, 14). The low level of sucrose remaining at the end of the night and the high rate of sucrose synthesis at the start of the day are consistent. Gradual inhibition of the sucrose synthesis pathway, perhaps from accumulation of cytoplasmic sucrose or other effectors, would lower sucrose synthesis to the steady state observed during the remainder of the day. At that point, allocation of triose phosphate to starch synthesis apparently came under close endogenous control. Near the end of the day, control shifted and the rate of starch synthesis responded more directly to the rate at which photosynthesis supplied triose phosphate.

To explore these hypotheses concerning mechanisms and priorities, several tasks remain. Sucrose phosphate synthetase activity has been found to vary in a manner consistent with a key role in controlling carbohydrate metabolism and translocation (8). It is important to measure it along with the levels of sucrose and other key metabolites in a range of types of plants in which detailed translocation and carbon allocation data are being taken. The stability of the starch synthesis rate needs to be examined under a variety of conditions. Control metabolite levels should be studied to determine which ones remain steady along with starch synthesis rate when conditions are changed. These effectors also should be measured as this control relaxes and starch accumulation slows at the end of the day.

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LITERATURE CITED

1. FONDY BR, DR GEIGER 1980 Effects of rapid changes in sink-source ratio on export and distribution of products of photosynthesis in leaves of *Beta vulgaris*, L and *Phaseolus vulgaris*, L. Plant Physiol 66: 945–949
2. FONDY BR, DR GEIGER 1982 Diurnal pattern of carbohydrate metabolism in source leaves of *Beta vulgaris* L. Plant Physiol 70: 671–676
3. FOX TC, DR GEIGER 1984 Effects of decreased net carbon exchange on carbohydrate metabolism in sugar beet source leaves. Plant Physiol 76: 763–768
4. GEIGER DR, B PLOEGER, TC FOX, BR FONDY 1983 Sources of sucrose translocated from illuminated sugar beet source leaves. Plant Physiol 72: 964–970
5. GEIGER DR, RT GIAQUINTA 1982 Translocation of photosynthate. *In* Govindjee, ed, Photosynthesis: Development, Carbon Metabolism, and Plant Productivity, Vol II. Academic Press, New York, pp 345–386
6. GORDON AJ, GJA RYLE, DF MITCHELL, CE POWELL 1982 The dynamics of carbon supply from leaves of barley plants grown in long or short days. J Exp Bot 133: 241–250
7. HUBER SC 1983 Relation between photosynthetic starch formation and dry-weight partitioning between the shoot and root. Can J Bot 61: 2709–2716
8. HUBER SC, DW ISRAEL 1982 Biochemical basis for partitioning of photosynthetically fixed carbon between starch and sucrose in soybean (*Glycine max Merr.*) leaves. Plant Physiol 69: 691–696
9. KAISER G, E MARTINOIA, A WIEMKEN 1982 Rapid appearance of photosynthetic products in the vacuoles isolated from barley mesophyll protoplasts by a new fast method. Z Pflanzenphysiol 17: 103–113
10. OUTLAW WH JR, J MANCHESTER 1979 Guard cell starch concentration quantitatively related to stomatal aperture. Plant Physiol 64: 79–82
11. REDGWELL RJ 1980 Fractionation of plant extracts using ion-exchange sephadex. Anal Biochem 107: 44–50
12. SALERNO GL, HG PONTIS 1978 Sucrose phosphate synthetase: separation from sucrose synthetase and a study of its properties. Planta 142: 41–48
13. STITT M, B HERZOG, HW HELDT 1984 Control of photosynthetic sucrose synthesis by fructose 2,6-bisphosphate. I. Coordination of CO_2 fixation and sucrose synthesis. Plant Physiol 75: 548–553
14. STITT M, B KURZEL, HW HELDT 1984 Control of photosynthetic sucrose synthesis by fructose 2,6-bisphosphate. II. Partitioning between sucrose and starch. Plant Physiol 75: 554–560