Effect of N-Source on Soybean Leaf Sucrose Phosphate Synthase, Starch Formation, and Whole Plant Growth[1]

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ABSTRACT

Soybeans (*Glycine max* L. Merr. cv Tracy and Ransom) were grown under N_2-dependent or NO_3^--supplied conditions, and the partitioning of photosynthate and dry matter was characterized. Although no treatment effects on photosynthetic rates were observed, NO_3^--supplied plants in both cultivars had lower starch accumulation rates than N_2-dependent plants. Leaf extracts of NO_3^--supplied plants had higher activities of sucrose phosphate synthase (SPS) and cytoplasmic fructose-1,6-bisphosphatase (FBPase) than N_2-dependent plants. The variation in starch accumulation was correlated negatively with the activity of SPS, but not the activity of FBPase, UDP-glucose pyrophosphorylase, or ADP-glucose pyrophosphorylase. These results suggested that starch accumulation is biochemically controlled, in part, by the activity of SPS. Leaf starch content at the beginning of the photoperiod was lower in NO_3^--supplied plants than N_2-dependent plants in both cultivars which suggested that net starch utilization as well as accumulation was affected by N source.

Total dry matter accumulation and dry matter distribution was affected by N source in both cultivars, but the cultivars differed in how dry matter was partitioned between the shoot and root as well as within the shoot. The activity of SPS was correlated positively with total dry matter accumulation which suggested that SPS activity is related to plant growth rate. The results suggested that photosynthate partitioning is an important but not an exclusive factor which determines whole plant dry matter distribution.

Understanding how photosynthate partitioning is regulated is of present interest because of the possible relationship between photosynthate partitioning and plant growth. Starch and sucrose are the primary end products of photosynthesis in many species and the synthesis requires the coordination of chloroplastic and cytoplasmic events (2, 15) linked via the chloroplastic phosphate translocator (10).

The pathways of sucrose and starch formation are interdependent as they compete for the pool of triose phosphates produced by the Calvin cycle. A consequence of this complex interdependence is that sucrose formation in the cytoplasm and starch formation in the chloroplast are reciprocally related (12). Thus, it appears that factors which regulate sucrose formation can also influence starch formation.

The biochemical regulation of sucrose biosynthesis is poorly understood, but, of enzymes involved in sucrose biosynthesis, two are presently recognized to be important in the regulation of the pathway. They are cytoplasmic FBPase,[2] EC 3.1.3.11 and SPS, EC 2.4.1.14 (1, 5, 7, 9, 21). The postulate that SPS and cytoplasmic FBPase are key regulatory enzymes of sucrose biosynthesis is based largely on kinetic properties exhibited by the isolated enzymes (1, 5, 9). Studies with soybeans have indicated that the activity of SPS in leaf extracts was inversely related to leaf starch content (12) and provide physiological evidence which supports the hypothesis that sucrose formation may be an important factor in regulating starch synthesis (19). Thus, SPS activity could be an important factor that may indirectly regulate starch formation (11). The activity of other enzymes of sucrose or starch formation were not measured, so their role in the regulation of photosynthate partitioning is not clear.

The relationship between photosynthate partitioning and plant growth is not fully understood, but studies with soybeans have indicated that increased starch accumulation is associated with increased shoot:root ratios (4). Thus, the partitioning of carbon during photosynthesis could be related to the partitioning of dry matter within the plant, which has been shown to be affected by genetic (8), environmental (4, 20), and nutritional (3, 18) factors. These factors could affect photosynthate partitioning as well as dry matter partitioning.

In separate studies with soybeans, the partitioning of whole plant dry matter (18) and the partitioning of carbon between starch and sucrose in mesophyll cells (12) have been shown to be affected by N source. Consequently, we have used two soybean genotypes, grown under different N regimes, as a system for studying the regulation of photosynthate partitioning and its relationship to plant growth. Our specific objectives were: (a) to characterize both photosynthetic starch formation and whole plant dry matter distribution, and (b) to determine whether differences in the partitioning of photosynthetically fixed CO_2 into starch were correlated with the activity of certain enzymes involved in starch and sucrose biosynthesis.

MATERIALS AND METHODS

Plant Culture. Soybeans (*Glycine max* [L.] Merr., cv Ransom and Tracy) were grown under nodulating or nonnodulating conditions. Seeds were germinated at 30°C and 95% RH for 3 d; those to be cultured under nodulating conditions were inoculated with *Rhizobium japonicum* (strain 3I1b110 or 3I1b31) and received N-free nutrient solution identical to that previously de-

[2] Abbreviations: FBPase, fructose-1,6-bisphosphatase; SPS, sucrose phosphate synthase; ADPG, adenosine 5'-diphosphoglucose; UDPG, uridine 5'-diphosphoglucose; CER, carbon dioxide exchange rate; GSH, reduced glutathione; PEG-20, polyethylene glycol-20000; Glc equiv, Glucose equivalent.

scribed (14), except that the phosphate concentration was 0.16 mM. The remaining plants were supplied with nutrient solution which contained 20 mM KNO₃. Potassium concentrations in the two solutions were equalized by supplementing the N-free solution with 10 mM K₂SO₄. The pH of both solutions was 6.0. The plants were grown for 62 to 65 d (see below) in a greenhouse between January and April in which natural light was supplemented for 14 h with metal halide lamps, which provided 550 $\mu E/m^2 \cdot s$ PAR at the bench top. Each of the six treatments consisted of four plants arranged in a randomized complete-block design with four replicates per treatment. At 62 (reps 1 and 2) and 64 (reps 3 and 4) d after planting, plants were assayed and harvested as described below.

Sampling Protocol and Net Photosynthesis. At 0800 and 1600 h, six 0.5 cm² punches were removed from the most recently, fully-expanded trifoliate leaf on the main axis of two plants and placed in 80% ethanol at −20°C until analyzed for starch as described below. During the interval in which starch accumulation was measured, net CER was measured on the same trifoliates using a Beckman[3] model 865 differential IR gas analyzer equipped with a 5-cm² plexiglass clamp-on cuvette. Air at the same temperature and CO₂ at the same concentration as in the ambient air was passed through the cuvette at a flow rate of 1.5 l/min and differences between CO₂ concentration in incoming and exhaust air streams were compared with standard CO₂ gases to determine CER. Exchange rates were expressed as mg CO₂ fixed/dm²·h⁻¹. Immediately after the punches were harvested at 1600 h, the trifoliates were excised, fresh weights and leaf areas determined, and the leaves were frozen at −80°C until assayed as described below.

Plant Harvest. On the following day (63 d for reps 1 and 2, 65 d for reps 3 and 4), the shoot was excised and divided into leaves, stems, and petioles. Xylem exudate was collected and analyzed for ureide-N and total-N as previously described (14) in order to assess the effectiveness of the imposed N-treatments. Mean xylem sap ureide-N expressed as a per cent of total-N was 8.8 and 14.4% in NO₃⁻-supplied plants of Tracy and Ransom, respectively. Ureide-N levels of nodulated plants of both cultivars were greater than 70% of the total-N. After collecting the xylem exudate, the roots were washed free of Perlite and then all the plant material was dried at 70°C for 72 h and weighed.

Starch Analysis. The leaf disks were ground in a mortar and pestle with 80% ethanol. The insoluble material was collected by centrifugation and washed again with hot 80% ethanol. Following centrifugation, the supernatant was decanted and the insoluble material was resuspended in 1.0 ml of 0.2 M KOH and boiled for 30 min. After cooling, the pH of the mixture was adjusted with 1 M acetic acid to about 5.5. Amyloglucosidase from *Aspergillus oryzae* (Sigma Chemical, Grade V) was used to digest the gelatinized starch. Preliminary investigations indicated that this preparation did not contain β-glucanase activity and quantitatively liberated glucose from amylopectin when used as described. Prior to use, 20 ml of the amyloglucosidase preparation was dialyzed against 2 L of 50 mM acetic acid-NaOH (pH 4.5) for 24 h with one change of buffer. The resulting solution was centrifuged to remove precipitated material and diluted to about 36 U/ml with the same buffer that was used for dialysis. One ml of the amyloglucosidase solution was added to each tube and then the samples were incubated for 30 min at 55°C. Following digestion, the samples were boiled for 1 min and all insoluble material was pelleted by centrifugation. An aliquot of the supernatant was then enzymatically assayed for glucose using hexo-

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[3] Mention of a trademark or proprietary product does not constitute a guarantee or warranty of the product by the United States Department of Agriculture and does not imply its approval to the exclusion of other products that may also be suitable.

kinase/glucose-6-P dehydrogenase as previously described (13).

Enzyme Extraction and Assays. Leaf tissue was extracted with buffer (8 ml/g fresh weight) for 30 s using a Brinkman Polytron homogenizer. The grind medium contained 50 mM Hepes-NaOH (pH 7.5), 5 mM MgCl₂, 1 mM EDTA, 2% (w/v) PEG-20, 1% (w/v) BSA, and 2 mM GSH. The brei was filtered through 8 layers of cheesecloth and any remaining intact cells were disrupted by passing the extract through a French pressure cell (330 kg/cm²). Debris was pelleted by centrifugation at 38,000g for 10 min. An aliquot of the supernatant was removed and used for ADPG pyrophosphorylase and UDPG pyrophosphorylase assays. Preliminary experiments indicated that soybean leaf ADPG pyrophosphorylase activity was strongly inhibited by DTT, but not by GSH, so the initial extract was prepared with GSH as described above. To the remaining supernatant, DTT was added to give a final concentration of 5 mM and the resulting mixture was used for SPS and cytoplasmic FBPase assays.

SPS was assayed by measuring fructose-6-P dependent sucrose-P (+ sucrose) formation from UDPG. The assay mixture (70 μl) contained 7.5 mM UDPG, 7.5 mM fructose-6-P, 15 mM MgCl₂, 50 mM Hepes-NaOH (pH 7.5), and an aliquot of leaf extract. Mixtures were incubated at 25°C for 10 min and were terminated by addition of 70 μl 1.0 N NaOH. Unreacted fructose-6-P was destroyed by heating the mixtures in a boiling waterbath for 10 min. After cooling, 0.25 ml of 0.1% (w/v) resorcinol in 95% ethanol and 0.75 ml of 30% HCl were added, and the mixture was incubated at 80°C for 8 min (16). The tubes were cooled to room temperature and the A_{520} was measured.

Cytoplasmic FBPase was assayed spectrophotometrically by measuring fructose-1,6-bisphosphate-dependent fructose-6-P formation under conditions which substantially inhibited chloroplast FBPase activity. The assay mixture (1.0 ml) contained 50 mM Hepes-NaOH (pH 7.0), 5 mM MgCl₂, 0.1 mM FBP, 0.2 mM NADP⁺, 2 units each of phosphoglucoisomerase and glucose-6-phosphate dehydrogenase, 1 unit of gluconate 6-P dehydrogenase, and an aliquot of leaf extract. Production of NADPH was monitored at 340 nm and 25°C.

UDPG pyrophosphorylase was assayed by measuring PPi-dependent glucose-1-P production using a continuous spectrophotometric procedure. The assay mixture (1.0 ml) contained 50 mM Hepes-NaOH (pH 7.5), 5 mM MgCl₂, 5 mM UDPG, 2 mM PPi, 15 units phosphoglucoisomerase, 5 units glucose-6-P dehydrogenase, 1 unit gluconate 6-P dehydrogenase, and an aliquot of leaf extract. The reaction was initiated with the leaf extract and the production of NADPH was monitored at 340 nm and 25°C.

ADPG pyrophosphorylase was assayed by measuring PPi dependent glucose-1-phosphate production using a continuous spectrophotometric assay. The assay mixture (1.0 ml) contained 50 mM Hepes-NaOH (pH 8.0), 5 mM MgCl₂, 1.0 mM ADPG, 2 mM PPi, 0.5 mM glycerate-3P, 0.3 mM NADP⁺, 15 units phosphoglucomutase, 5 units glucose-6-phosphate dehydrogenase, 1 unit gluconate 6-P dehydrogenase, and an aliquot of leaf extract. The reaction was initiated with the leaf extract and the production of NADPH was monitored at 340 nm and 25°C. At the concentration of ADPG indicated, glycerate-3P was added to give maximum activation. The concentration of ADPG used in the assay gave activities 50% of maximum velocity, but cost of the substrate precluded assaying the enzyme under saturating conditions.

RESULTS

Photosynthesis and Starch Formation. There were no significant treatment effects on net CER of the most recently, fully expanded leaves on the main stem (Table I). Starch accumulation rates, as measured by the difference between 1600 and 0800 h starch contents, were normalized with net CER in order to take

Table I. *Effect of Soybean Cultivar and Nitrogen Source on net CER, Relative Starch Accumulation, and Morning Starch Content of the Most Recent Fully Expanded Trifoliate at 65 Days after Emergence*

The data represents mean ± SE of four replicates.

Cultivar	Nitrogen Source	Net CER	Relative Starch Accumulation[a]	Starch Content at 0800 h[a]
		$mg\ CO_2\ fixed$ $dm^2 \cdot h$	$mg\ Glc\ equiv$ $mg\ CO_2\ fixed$	$mg\ Glc\ equiv$ dm^2
Tracy	NO₃	20.4 ± 3.2	0.24 ± 0.04	45.5 ± 11.9
	Rj 110	18.2 ± 2.2	0.39 ± 0.05	66.3 ± 11.8
	Rj 31	18.3 ± 2.9	0.34 ± 0.05	62.8 ± 8.4
Ransom	NO₃	18.8 ± 2.4	0.21 ± 0.04	31.2 ± 5.8
	Rj 110	18.4 ± 0.7	0.28 ± 0.03	59.6 ± 3.9
	Rj 31	19.7 ± 1.9	0.31 ± 0.05	50.8 ± 5.8

[a] Analysis of variance showed significant ($P \leq 0.05$) N source effect.

into account the differences in net CER between plants. The ratio of starch accumulation to CER, hereafter referred to as relative leaf starch accumulation, provides an estimate of the partitioning of carbon into starch. In both cultivars, plants fed supplemental NO_3^- had the lowest rates of relative starch accumulation. Differences between strains within cultivars as well as differences between cultivars were not significant. Starch content at 0800 h was lowest in NO_3^--supplied plants in both cultivars, but differences between strains within cultivars and between cultivars were not significant. Starch content at 0800 h is an estimate of the difference between accumulation and mobilization of starch. The results obtained suggested that N_2-dependent plants were unable to mobilize the starch accumulated during the photosynthetic period to the same extent as NO_3^--supplied plants.

Activities of Sucrose and Starch Biosynthetic Enzymes. The activities of two enzymes required for sucrose formation were affected by the imposed treatments (Table II). The activity of SPS in leaf extracts was affected significantly by N-nutrition in both cultivars as SPS activity was higher in NO_3^--supplied plants than N_2-dependent plants. Strain effects were not significant in either cultivar, but SPS activity was higher in cultivar Tracy for all N-treatments than in cultivar Ransom. Cytoplasmic FBPase activity was higher in NO_3^--supplied plants compared to plants dependent on N_2-fixation in both cultivars. Greater treatment differences in the activity of cytoplasmic FBPase were observed in cultivar Tracy and, therefore, a significant (0.05 level) cultivar × N source interaction was observed. The activities of UDPG pyrophosphorylase and ADPG pyrophosphorylase were not affected by the imposed treatments in the present study.

The partitioning of photosynthate into starch was correlated

negatively with SPS activity in leaf extracts of both cultivars ($r = -0.78$, -0.72; Fig. 1). Even though treatment effects on cytoplasmic FBPase activity were observed, there was no significant correlation with starch accumulation ($r = -0.43$, -0.34; Fig. 2). Similarly, no correlation between starch formation and activities of UDPG or ADPG pyrophosphorylase was observed (data not shown). The results indicate that alterations in the partitioning of photosynthate into starch are not associated with changes in the activities of all of the enzymes of sucrose or starch formation, but only with changes in the activity of SPS (19).

Leaf N Concentration, Plant Growth, and Dry Matter Partitioning. Leaf total N concentration was affected by the imposed treatments in both cultivars (Table III). Leaves of NO_3^--supplied plants had the highest total N concentration. Leaf total N concentration in plants inoculated with *Rhizobium japonicum* strain 110 was similar to that in NO_3^--supplied plants, whereas N concentration in leaves of plants inoculated with *Rhizobium japonicum* strain 31 was from 12 to 27% lower than that in leaves of NO_3^--supplied plants. Total dry matter accumulation and its partitioning among plant parts were affected by N-source in both cultivars. Plants supplied NO_3^- accumulated more total dry matter than plants inoculated with either *Rhizobium* strain. Plants in Tracy inoculated with strain 110 accumulated more dry matter than those inoculated with strain 31; differences in total dry matter between strains with Ransom were not significant at the 0.05 level. Total dry matter accumulation was not significantly different in the two cultivars.

Total dry matter accumulation and SPS activity were affected by N-source in both cultivars. As a result, the activity of SPS in leaf extracts was correlated positively with total dry matter accumulation in both cultivars ($r = 0.80$, $r = 0.78$; Fig. 3). These results suggested that SPS activity was correlated with growth rate in these cultivars.

Although total dry matter accumulation was similar in Ransom and Tracy, the two cultivars differed in how dry matter was partitioned within the plant (Table III). Cultivar and N-source affected shoot:root ratios which were higher for Tracy as compared to Ransom for all N-treatments. Plants supplied NO_3^- had the highest shoot:root ratios in both cultivars. Not only were differences in dry matter distribution between the shoot and root observed, but also differences in the partitioning of dry matter within the shoot. Leaf:(stem + petiole) ratios were higher in Tracy than in Ransom for all N-treatments and with Tracy, plants supplied NO_3^- had higher leaf:(stem + petiole) ratios than plants inoculated with either *Rhizobium* strain. Leaf:(stem + petiole) ratios were not affected by N-source in Ransom. Together, these results suggested that soybeans grown under these conditions responded to supplemental NO_3^-, in part, by increased shoot:root ratios, but in one cultivar (Tracy) the increase

Table II. *Effect of Soybean Cultivar and Nitrogen Source on the Activities of Certain Enzymes Involved in Sucrose or Starch Biosynthesis in the Most Recent Fully Expanded Trifoliate at 65 Days After Emergence*

The data represent mean ± SE of four replicates.

Cultivar	Nitrogen Source	Sucrose Phosphate Synthase[a]	Cytoplasmic FBPase[b]	UDPG Pyrosphosphorylase	ADPG Pyrophosphorylase
			$\mu mol\ product/dm^2 \cdot h^{-1}$		
Tracy	NO₃⁻	33.9 ± 2.9	38.7 ± 1.7	173 ± 34	69 ± 5
	Rj 110	21.4 ± 2.7	29.0 ± 4.6	169 ± 41	77 ± 11
	Rj 31	18.0 ± 3.7	21.3 ± 0.7	167 ± 37	74 ± 10
Ransom	NO₃⁻	20.2 ± 3.0	34.5 ± 1.5	95 ± 6	55 ± 8
	Rj 110	15.2 ± 1.2	29.3 ± 1.6	102 ± 20	60 ± 3
	Rj 31	13.0 ± 1.9	30.0 ± 2.8	131 ± 35	66 ± 6

[a] Analysis of variance showed significant ($P \leq 0.01$) N-source and cultivar effects.
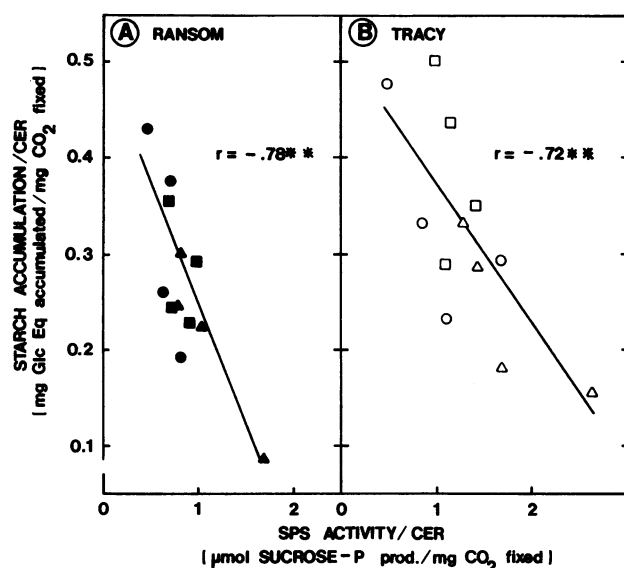[b] Analysis of variance showed significant ($P \leq 0.05$) N source × cultivar interaction.

FIG. 1. Leaf starch accumulation compared with activity of SPS in leaf extracts of two soybean varieties grown under NO_3^--supplied (▲, △) or N_2-dependent conditions (●, ○ strain 31; ■, □ strain 110). Closed and open symbols represent cultivars Ransom and Tracy, respectively. Both parameters have been normalized by CER in order to account for small differences in photosynthetic rates (**), Indicates that the correlation coefficient was significant at the 1% level.
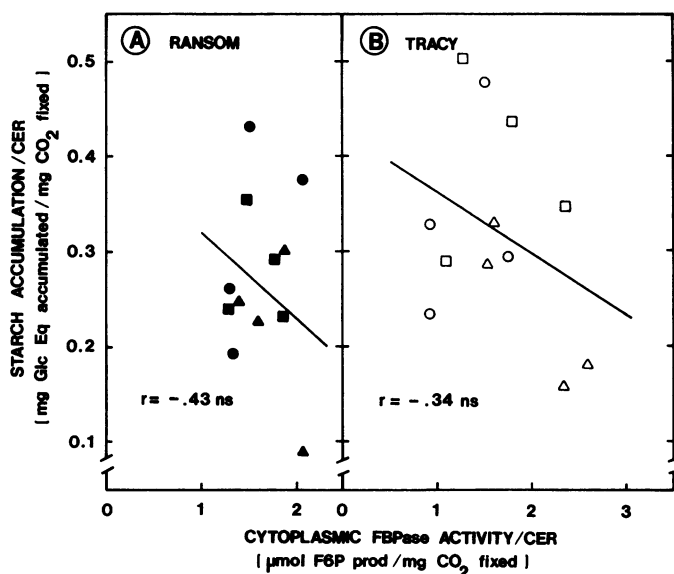

FIG. 2. Leaf starch accumulation compared with activity of cytoplasmic FBPase in leaf extracts of two soybean varieties grown under NO_3^--supplied (▲, △) or N_2-dependent conditions (●, ○ strain 31; ■, □ strain 110). Closed and open symbols represent cultivars Ransom and Tracy, respectively. Both parameters have been normalized by CER in order to account for small differences in photosynthetic rates.

in shoot growth relative to root growth was predominately the result of increased leaf growth, whereas the other cultivar (Ransom) responded with essentially equal increases in leaf and stem growth relative to root growth.

DISCUSSION

Biochemical Regulation of Photosynthate Partitioning. The partitioning of photosynthetically fixed carbon into starch has been suggested to be biochemically controlled, in part, by the

rate of sucrose formation (12, 19). The results of the present study are consistent with this postulate as the rate of starch accumulation was inversely related to the activity of SPS in leaf extracts (Table I; Fig. 1). The activities of some other enzymes of sucrose synthesis (cytoplasmic FBPase, UDPG pyrophosphorylase) or starch synthesis (ADPG pyrophosphorylase) were either not affected by the imposed treatments or not significantly correlated with starch accumulation rates. This suggests that the imposed treatments resulted in specific effects on enzyme activity rather than affecting the enzymes of sucrose and starch biosynthesis in a concerted manner. These results are consistent with a previous study in which the activity of SPS but not ADPG pyrophosphorylase was affected by irradiance treatments (19) and suggest that environmental and nutritional factors can affect photosynthate partitioning by a similar mechanism. The results of the present study are not suitable for assessing the relative importance of alterations in total enzyme activity *versus* metabolic regulation *in vivo* to the control of photosynthate partitioning. Conceivably, both processes are essential components that work in concert to regulate carbon partitioning.

SPS Activity and Plant Growth Rate. Recent evidence suggests that the activity of SPS in soybean leaves is affected by source:sink alterations (17). When the demand for sucrose was enhanced by partial source leaf defoliation, increased activities of SPS were observed. Results from the present study suggest that a similar relationship exists *in situ* as SPS activity was correlated positively with total dry matter accumulation in both cultivars. In other experiments, we have observed that differences in total dry matter of NO_3^--supplied *versus* N_2-dependent plants reflect differences in relative growth rate (unpublished results). Because SPS activity was correlated positively with total dry matter accumulation (Fig. 3), the activity of SPS appears to be closely associated with plant growth.

Nutritional Effects on Photosynthate Partitioning and SPS Activity. Previously, it was reported that the partitioning of photosynthetically fixed carbon into starch was greater in mesophyll cells isolated from leaves of NO_3^--supplied plants than cells from nodulated plants (12). In the present study, nodulated plants had higher rates of starch accumulation and lower levels of SPS activity than NO_3^--supplied plants. At present, the cause of this discrepancy is not clear. Leaves from plants in reproductive growth were used to isolate cells in the previous study, while nonflowering plants were used in the present study. In addition, the isolated cells are not in the same environment that exists *in vivo* and conceivably these factors could influence the manner in which N nutrition affects the partitioning of carbon between end products.

It is apparent that differences in leaf N concentration cannot fully explain the observed differences in SPS activity and photosynthate partitioning. For example, SPS activity was higher in NO_3^--supplied plants compared to N_2-dependent plants inoculated with *R. japonicum* strain 110 but leaf N concentration was similar in these plants. In addition, leaf N concentration was higher in N_2-dependent plants inoculated with strain 110 compared to that in plants inoculated with strain 31, whereas differences in SPS activity between the two strains were not statistically significant. Collectively, these results suggested that N source rather than leaf N concentration *per se*, may affect the activity of SPS and photosynthate partitioning. Since dry matter accumulation as well as photosynthate partitioning was affected by the source of N nutrition, it was not possible to determine whether differences in starch accumulation and enzyme activities were due to a direct influence of the source of N nutrition or due to an influence of growth rate which resulted from the imposed N treatments. Regardless of the mechanism by which N source influenced partitioning, results from the present study suggest that photosynthate was inefficiently partitioned in

Table III. *Effect of Soybean Cultivar and Nitrogen Source on Leaf Nitrogen Concentration, Whole Plant Dry Matter Accumulation, and Dry Matter Partitioning at 65 Days after Emergence*

The data represent mean ± SE of four replicates.

| Cultivar	Nitrogen Source	Leaf N Concentration[a]	Total Dry Matter[b]	Shoot wt Root wt[a]	Leaves Stems + Petioles[a]
		%	g/plant		
Tracy	NO$_3$⁻	3.99 ± 0.05	47.1 ± 2.8	5.62 ± 0.44	1.43 ± 0.10
	Rj 110	3.68 ± 0.11	37.9 ± 2.1	4.70 ± 0.24	1.20 ± 0.03
	Rj 31	3.14 ± 0.08	27.6 ± 1.0	4.58 ± 0.33	1.28 ± 0.04
Ransom	NO$_3$⁻	4.00 ± 0.09	45.4 ± 2.6	4.84 ± 0.32	0.96 ± 0.02
	Rj 110	3.86 ± 0.05	36.7 ± 2.7	4.17 ± 0.18	0.98 ± 0.03
	Rj 31	3.56 ± 0.06	33.5 ± 1.7	3.65 ± 0.34	0.98 ± 0.02

[a] Analysis of variance showed significant (P ≤ 0.05) N source and cultivar effects.
[b] Analysis of variance showed significant (P ≤ 0.005) N source effect.
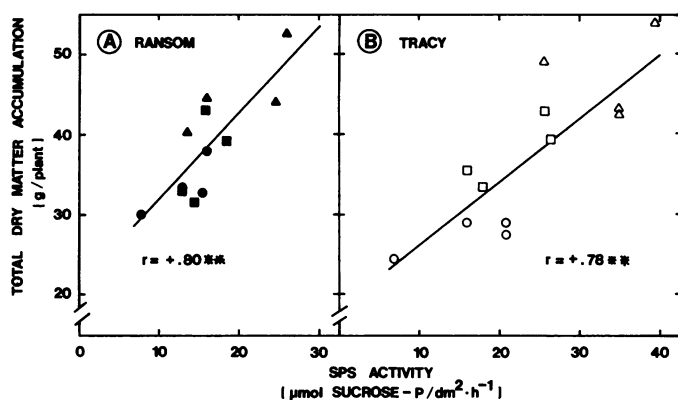
FIG. 3. Total plant dry matter accumulation compared with SPS activity in leaf extracts of two soybean varieties grown under NO$_3$⁻-supplied (▲, △) or N$_2$-dependent conditions (●, ○ strain 31; ■, □ strain 110). Closed and open symbols represent cultivars Ransom and Tracy, respectively. (**), Indicates that the correlation coefficient was significant at the 1% level.

nodulated plants and are consistent with the suggestion by Finn and Brun (6) that nodule activity is limited by an inefficient partitioning and utilization of photosynthate rather than by current photosynthesis.

Relationship between Photosynthate Partitioning and Plant Growth. The relationship between the partitioning and utilization of photosynthate and whole plant growth is complex and poorly understood. Increased starch accumulation in leaves of nonnodulated soybeans due to changes in the photosynthetic period has been associated with increased shoot growth relative to root growth (4). Thus, carbon mobilized during the dark presumably was retained primarily in the shoot. In the present study, the accumulation and net utilization of starch were affected by N-nutrition. Data pooled from both cultivars (because cultivar effects on relative starch accumulation and morning starch content were not significant) indicated that N-nutrition affected morning starch levels to a greater extent than relative starch accumulation. This suggests that although N$_2$-dependent plants partitioned more of their photosynthate into starch than their NO$_3$⁻-supplied counterparts, comparatively less carbon was made available for growth and respiration at night. Since nodulated plants had lower shoot:root ratios than NO$_3$⁻-supplied plants, the results of this study are, in part, consistent with the postulate (4) that the partitioning of carbon into starch can influence the distribution of dry matter within the plant. However, this study indicates that factors other than photosynthate partitioning/utilization are also involved in determining the partitioning of dry matter within the plant. This is evident from the

observation that although the two cultivars differed in the manner in which dry matter was partitioned within the shoot and plant, no statistical differences in photosynthate partitioning between cultivars were present. It is not surprising that measurements of a single leaf do not fully explain whole plant growth, as the sampled leaf was but a small component of the photosynthetic canopy and was observed at one period during its development. Nonetheless, our results suggest that photosynthate partitioning is an important factor related to plant growth and it appears to be biochemically regulated, in part, by the activity of SPS. Additional information about how genetic and nutritional factors influence the partitioning and utilization of photosynthate throughout the canopy and over time would provide a more complete understanding of the relationship between partitioning and plant growth.

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