Plant Physiol. (1980) 65, 975-979
0032-0889/80/65/0975/05/$00.50/0

Kinetics of ^{14}C-Photosynthate Uptake by Developing Soybean Fruit

Received for publication September 28, 1979 and in revised form January 2, 1980

JOHN H. THORNE
Department of Ecology and Climatology, The Connecticut Agricultural Experiment Station, New Haven, Connecticut 06504

ABSTRACT

By pulse-labeling field-grown soybean leaves for 60 seconds at midday with $^{14}CO_2$ and then sequentially harvesting, dissecting, and extracting the radioactive fruit tissues (of pod and seeds), the route, uptake kinetics, and metabolic fate of ^{14}C-photosynthate as it was imported by 35- to 40-day-old pods were determined. As the [^{14}C]sucrose pulse entered the pods, the seeds became radioactive immediately but a lag of nearly 30 minutes occurred before label could be detected in the pod wall pericarp.

Import of the ^{14}C-pulse by the seeds was exclusively via the seed coats, where rapid unloading occurred. Maximum accumulation of label in the seed coat occurred in about 60 minutes at which time 59% of the total radioactivity in the fruit was in the three seed coats, whereas only 7% was in the cotyledons they enclosed. The photosynthate remained as [^{14}C]-sucrose as it passed through the seed coat, but appeared to be hydrolyzed relatively soon after uptake by the cotyledons. By 2.5 hours, 60% of the ^{14}C-photosynthate pulse had passed into the cotyledons with only 27% remaining in the seed coats. Inasmuch as there is no vascular connection between maternal seed coat and the developing embryo, cotyledonary uptake of sucrose released from the inner seed coat surface may require specialized transport mechanisms.

In the very early stages of soybean seed growth, nourishment is obtained indirectly through extensive digestion of maternal ovule tissues. During this period the seeds enlarge slowly whereas the surrounding pod (ovary) grows rapidly (3). Soon, however, seeds utilize external photosynthate supplied via the pod. Biochemical studies of oil and protein deposition during soybean seed growth (1) indicate that large quantities of photosynthate are transported to the embryo and cotyledons as they develop. Import of photosynthate necessary to maintain reported (4) rates of seed growth (3–8 mg dry weight seed^{-1} day^{-1}; [4]) can be as much as 7–20 mg sucrose seed^{-1} day^{-1} (calculated from ref. 14), approximately half of the sucrose production of each dm^2 of illuminated leaf per day. Since the upper, more fully-illuminated leaves commonly supply several pods, some subtending and some at other nodes, photosynthate availability may at times limit seed growth and productivity. Thus, although concurrent canopy photosynthesis is usually the primary source of photosynthate for seed growth (12, 15), redistribution of stored assimilates from pod walls (25) and stems (24) may contribute substantially in some cultivars.

Anatomical studies on developing seeds in the Graminae have found specialized absorption structures in the region of seed attachment, where a discontinuity of phloem exists (27). Biochemical studies of the kinetics and mechanism of ^{14}C-photosynthate uptake, however, leave considerable debate as to whether sucrose can be transported from the phloem into the developing endosperm in an unmodified form (16, 22). A similar lack of vascular continuity is known to exist between embryonic and maternal tissues of various legume species. including soybean (3, 20). While the literature contains studies of soybean phloem loading and transport kinetics (5, 23) and whole plant photosynthate distribution patterns (2), there is little information characterizing photosynthate uptake and metabolism by the reproductive sink tissues of soybean (18, 19, 25) and none regarding the mechanisms involved.

A thorough characterization of phloem unloading and photosynthate uptake by developing soybean seeds is necessary to understand the mechanism of long distance translocation, and to determine if photosynthate availability may sometimes limit seed growth and productivity. The route and kinetics of ^{14}C-photosynthate uptake by developing soybean seeds are reported here; short-term distribution and metabolism within fruit tissues are discussed, and findings are related to possible controls of photosynthate partitioning and seed productivity.

MATERIALS AND METHODS

Labeling Procedure. On selected plants within a population of field-grown Amsoy-71 soybeans (*Glycine max* (L.) Merr.), a single leaflet was photosynthetically labeled with $^{14}CO_2$ (290 μl l^{-1} CO_2, 1.1% O_2, 1.35 mCi mol^{-1} CO_2) shortly after solar noon on a clear day, approximately 35 days after flowering at that node. Radiant flux density (PAR), was 2.3×10^3 μE m^{-2} s^{-1} as measured with a Licor-185 sensor. To reduce variability, replicate plants were selected for labeling that met the following requirements: between mainstem nodes 12 and 14 there was a recently fully expanded leaf, west-facing for uniform afternoon solar exposure, with a 32-cm petiole, three subtending pods at that node, each with three seeds 35–40 days of age (as determined by an external pod width measurement with calipers), and which was at least 2 m removed from other experimental plants. A 40-cm^2 area in the center of the terminal leaflet was labeled for 60 s using a clip-on assimilation chamber built of Plexiglas with windows of IR-transparent polypropylene Propafilm C110 (Imperial Chemical Industries, Ltd., Wilmington, Del.). With this chamber both abaxial and adaxial leaf surfaces were simultaneously exposed to $^{14}CO_2$ supplied from a small lecture bottle. Plants were then allowed a "chase" period of varying lengths for photosynthesis and translocation.

Recovery and Analysis of ^{14}C-Photosynthate. Harvest intervals following labeling were chosen after a preliminary experiment with this system had identified the time of ^{14}C arrival in the fruit. Harvested plants from the preliminary experiment were quickly cut into appropriate sections and frozen in liquid N_2. Petioles were cut into 2-cm sections, frozen and later digested in 1.0 ml 70% $HClO_4$, 30% H_2O_2 (1:1, v/v) in capped scintillation vials at 60 C. The pods and seeds of this preliminary experiment were similarly digested. Digested samples were diluted with water and counted by liquid scintillation spectroscopy in Aquasol II cocktail (New

England Nuclear) using external standard correction.

Using the results of the preliminary experiment, appropriate fruits from three plants at each interval were harvested, quickly frozen on solid CO_2, and later the frozen tissues were dissected into pod wall pericarp, major pod vascular bundles (ventral and dorsal), seed coats, and cotyledons plus embryos. After freeze drying, tissues were carefully fractured and extracted with boiling 80% ethanol (v/v) in a microsoxhlet apparatus for 48 h. The ethanol-insoluble tissue was then digested as described above.

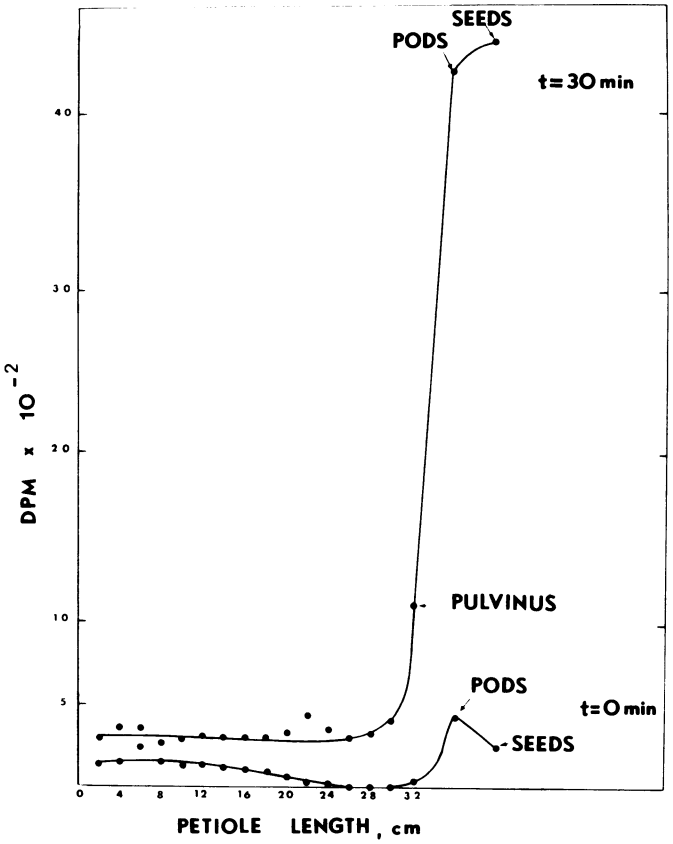

FIG. 1. Experimental basis for estimates of import velocity and time of arrival of ^{14}C-photosynthate in soybean fruit 31.8 ± 0.3 cm away from a leaflet exposed to $^{14}CO_2$. In this preliminary experiment, three plants were harvested every 15 min for 2 h after labeling and the radioactivity determined in 2-cm petiole lengths and fruit parts (seeds and pod walls, including major pod veins) by digestion and counting as described in the text. The two harvest periods illustrated (t = 0, t = 30) correspond to 30 and 60 min after exposing the leaf to $^{14}CO_2$ for 1 min.

Table I. *Partitioning of [^{14}C]Sucrose between Seeds and Pod Walls as It Arrives in the Fruit*

Each value represents the mean percentage distribution of total radioactivity among fruit components during uptake; three fruit at each time interval. Pod wall pericarp does not include the dorsal or two ventral bundles.

Time in Fruit	^{14}C Distribution	
	Seeds	Pod Wall Pericarp
min	%	
15	27.1	0
30	32.5	20.5
45	64.8	14.9
60	66.1	13.5
150	86.2	7.4

Ethanol-soluble photosynthate was dried at 35 C under reduced pressure. Lipids were extracted from the dried photosynthate with petroleum etherdiethyl ether (9:1, v/v), evaporated to about 0.2 ml under N_2, and radioactivity counted in Aquasol II.

The water-soluble residue was dissolved in 50% ethanol (v/v) and further fractionated on coupled, low pressure ion exchange columns. Following elution of the neutral sugar fraction with 40 ml H_2O, the columns were separated and an amino acid fraction (eluted from the Dowex 1 anion resin with 25 ml 2 N HCl) were collected. Aliquots of the basic and acidic fractions were counted in Aquasol II. The neutral fraction was dried at 35 C under reduced pressure, redissolved in 2.0 ml 50% ethanol (v/v), and the radioactivity in aliquots counted. Chemical analyses were as previously described (25). Sugars, separated and identified by TLC (25), were scraped from the plates into scintillation vials, eluted with 3 ml 50% ethanol (v/v), and counted as a thixotropic gel with

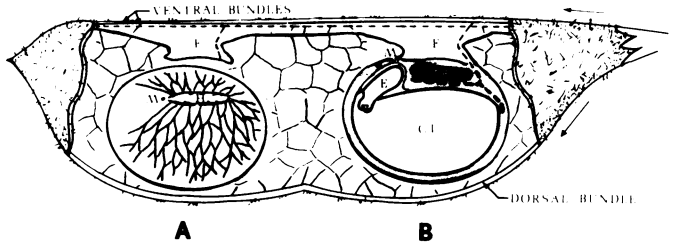

FIG. 2. Routes for distribution of photosynthate within developing soybean fruit. (A) surface view of a detached seed rotated about 70 degrees toward the viewer, illustrating the subsurface venation anastamosing repeatedly from two parallel lateral branches that lie just under the hilum. (B) sketch of a longitudinal section through a seed attached by the funiculus to the pod's ventral bundle. The single vascular bundle entering at the chalazal end of the hilum region branches below the tracheid bar. From one of these lateral branches a single vein of the anastamosing network can be seen to lie within the seed coat at right. F = funiculus, H = hilum, E = embryonic axis, M = micropyle, CT = cotyledon, TB = tracheid bar.

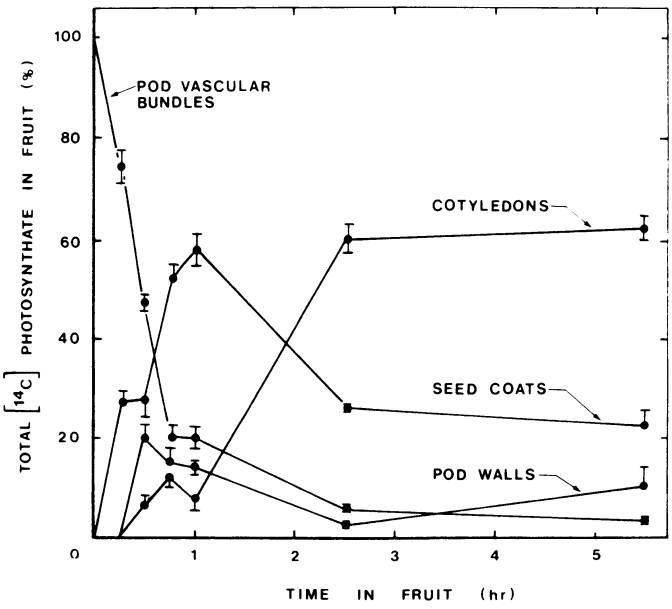

FIG. 3. Kinetics of ^{14}C-photosynthate uptake by soybean fruit. Illustrated is the distribution of label in tissues at various times after arrival in the fruit. Points represent the mean percentage distribution ± SE of radioactivity among fruit tissues during uptake; three fruit at each time interval.

Aquasol II.

Autoradiography. To examine the radial distribution of ^{14}C-photosynthate within seed coats, plants with seeds of several known ages were photosynthetically labeled with $^{14}CO_2$ for 5 min. After 90 min, freshly harvested seed coats were quickly washed in several changes of ice cold 1 mM $CaCl_2$, blotted dry, frozen in a flattened position, and freeze-dried. To obtain clear, reproducible autoradiographs, they were then further flattened by compressing at 25,000 p.s.i. in a hydraulic plant press, and autoradiographed for 1.5 weeks against Kodak Panatomic X film.

Microscopy. Anatomical observations were made with bright-field light microscopy on sectioned fruit tissues stained with 0.05% toluidine blue.

RESULTS

Kinetics of Transport to the Fruit. Before presenting data on the uptake of photosynthate by the fruit, the transport kinetics between a source leaf and fruit at that node should be considered. Export of ^{14}C-photosynthate from recently expanded leaves to subtending pods approximately 35–40 days after flowering at that node was quite rapid. Through sequential harvesting and extraction of petiole segments, movement of the radioactive front was observed to proceed at approximately 0.9 cm min^{-1}, including the time necessary for phloem loading in the leaf. Fractionation of the photosynthate extracts by TLC confirmed that sucrose was the predominant component. First accumulations of [^{14}C]sucrose in the fruit tissues were observed approximately 30 min after $^{14}CO_2$-pulsing the source leaf 32 cm away (Fig. 1). The pulvinus at the base of the petiole accumulated small amounts of ^{14}C as the pulse passed out of the petiole. Label could often be detected in the fruit tissue before the major [^{14}C]sucrose front had fully traversed the petiole, indicating a more rapid movement of small amounts of label, probably due to pulse broadening. Sucrose entering the pod is immediately divided between the two ventral bundles, along which the seeds are attached alternatively, and the dorsal vascular bundle. Extensive secondary vascular branching from these major bundles also supplies photosynthate to the pod wall pericarp for initial pod growth and later, for temporary assimilate storage in some cultivars (25). These secondary veins are imbedded in pod wall pericarp to varying depths, surfacing on the inside where the pericarp touches the seeds. Despite this extensive pod wall vascularization, the seeds are the primary sink for entering sucrose (Table I). As [^{14}C]sucrose traverses the length of the major pod vascular bundles, the attached seeds become radioactive immediately whereas no ^{14}C can be detected in the pod walls for approximately 30 min after arrival of the ^{14}C-front in the fruit (Table I). Accumulation in the seeds is rapid; by 45 min, more than 65% of the radioactivity in the fruit is in the seeds, 20% of the pod veins and 15% in the pod walls.

Anatomical Evaluation of Uptake Route. Anatomical observations of the fruit tissues involved in photosynthate import are summarized in Figure 2. Sucrose enters the seeds from the pod ventral bundles through the funiculus and the hilum region of the seed coat. The head of the funiculus is expanded into a bordered disc, which fits into the depression of the hilum. Through it passes a single secondary vascular bundle at the posterior end. Upon entering the seed coat this vascular bundle branches into two lateral bundles just below the thick tracheid bar (3, 11). Within the seed coat, these branch bundles parallel the hilum from the chalazal branch point to the micropyle. Along these two branch bundles, about 10–15 veins depart from each (Fig. 2), anastamosing repeatedly throughout the seed coat near the surface facing the cotyledons until it is extensively vascularized except around the micropyle (20). No vascular connection exists to carry photosynthate from the maternal seed coat to the developing embryo (and cotyledons) of the seeds. They are, in fact, separated by a thin transparent tissue, the embryo sac.

Kinetics of Sucrose Unloading in Seed Coats. The 60-s exposure of the leaf to $^{14}CO_2$ of high specific radioactivity produced a rather brief pulse of [^{14}C]sucrose moving into the seed. Figure 3 illustrates the kinetics of [^{14}C]sucrose uptake by the seeds. Confirming the anatomical observations, the primary route of import from a pod ventral bundle was found to be via the seed coats. Approximately 15 min after arrival of the ^{14}C-pulse in the fruit, 27% of the label was in the seed coats with the remaining 73% still in the major pod vascular bundles. Label did not appear in any other fruit tissue until after approximately 30 min when the pod walls contained 20.5%, and the cotyledons contained 6% of the total.

Accumulation of label in the seed coats continued almost linearly (Fig. 4) to a maximum at about 60 min after arrival in the fruit. At that time 59% of the total ^{14}C in the fruit was in the three seed coats—fully a 3-fold greater level than the supplying pod veins. This rapid accumulation of label occurred into an existing sucrose pool comprising approximately 7.4% of the seed coat dry weight (Table II). No metabolism of the [^{14}C]sucrose pulse was observed during passage through the seed coat, either prior to or after unloading of [^{14}C]sucrose from the vascular network within the seed coat (Table II).

Distribution of Label within Seed Coats. Autoradiography of seed coats (surface exposures) from plants exposed to $^{14}CO_2$ revealed the ^{14}C was not restricted to the seed coat venation, but was uniformly distributed across the flattened seed coats including the micropyle region. Autoradiographs of young seed coats lacking a developed venation (13 days after flowering) also revealed uniform label distribution across the surface. That the label is confined to the innermost cell layers of the seed coat is suggested by the location of the vascular tissue there and by autoradiography. When the outside surfaces of the seed coats were placed next to the film, considerably less film exposure occurred than when the inside surface was next to the film (data not shown).

Kinetics of Uptake by Cotyledons. Compared to the uptake of

Table II. *Seedcoat Characteristics of Seeds Harvested Approximately 60 min after Arrival of ^{14}C Photosynthate from Leaves of a $^{14}CO_2$-labeled Field-grown Amsoy-71 Soybean Plant*

Frozen fruit were dissected, seed coats removed, washed briefly in ice-cold 1 mM $CaCl_2$, and freeze dried. Extraction and determination of ^{14}C metabolites were as detailed in the text. Each entry is the mean of three seedcoats taken from seeds in the same pod, all pods from the same plant.

Dry Weight	Extractable with 80% Ethanol	H_2O	Sucrose	^{14}C as Sucrose	^{14}C as Reducing Sugars	Radioactivity
mg	*% dry wt*	*%*	*% dry wt*	*% of total*		*dpm × 10^{-3}*
17.1	47	73.6	7.8	97	1.0	6.7
18.0	42	74.2	7.5	98	0.9	7.5
18.7	44	74.8	6.9	93	0.9	1.3
17.9	44.3	74.2	7.4	96	0.9	$\bar{x}$

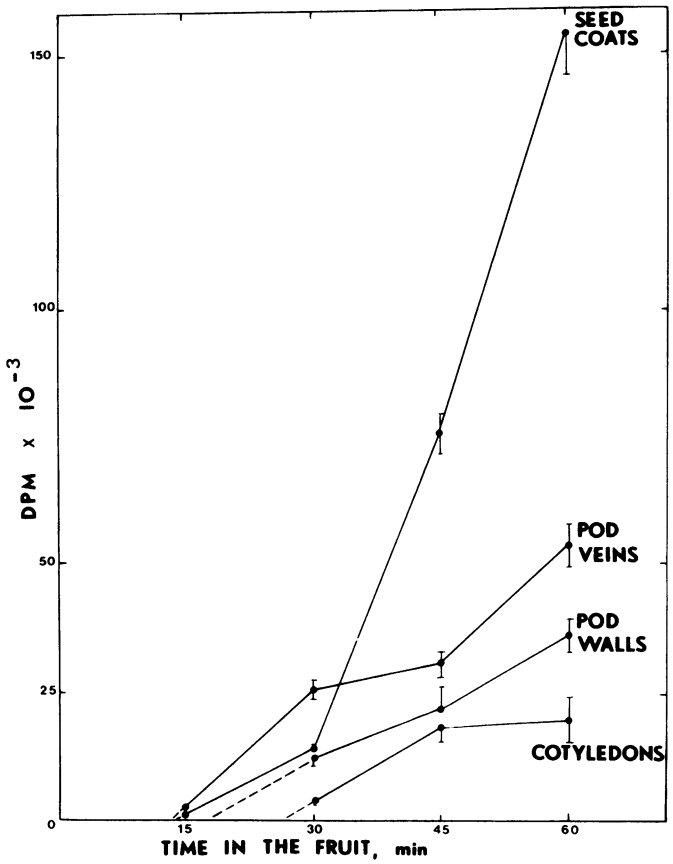

FIG. 4. Accumulation of ^{14}C-photosynthate in fruit tissues during the first 60 min after arrival of the ^{14}C pulse in the fruit. Points represent the mean dpm ± SE of radioactivity among fruit tissues during uptake; three fruit at each time interval.

Table III. *Distribution of ^{14}C among Photosynthate Fractions in Soybean Cotyledons 60 min after Arrival of the ^{14}C-Pulse in the Fruit*

Each value is the mean ± SE of nine cotyledon pairs.

Cotyledonary Fraction	^{14}C
	%
Neutral sugars	73.7 ± 3.1
Sucrose	26.7
Glucose	19.5
Fructose	17.6
Others	9.9
Amino acids	12.4 ± 1.3
Organic acids	12.3 ± 2.1
Insoluble	1.6 ± 0.8
Lipid	0
7.4 ± 1.7% total ^{14}C	100

[^{14}C]sucrose by the seed coats, label entered the cotyledons from the surrounding seed coat slowly (Fig. 3). During 60 min of nearly linear [^{14}C]sucrose accumulation in the seed coats, only small amounts of label appeared in the cotyledons (Fig. 4). Fractionation of the cotyledonary ^{14}C-sugars (only 5.8% of the total radioactivity in the fruit at this time) yielded predominantly [^{14}C]sucrose. Also measurable amounts of [^{14}C]glucose and [^{14}C]fructose were obtained (Table III), indicating that sucrose may be taken up intact and hydrolyzed thereafter. Once hydrolyzed, further metabolism of ^{14}C-photosynthate is rapid, for 25% of the label in the cotyledons at that time could be recovered from the basic (amino acid) and acidic (organic acid) fractions. Such metabolism of photosynthate was not evident in petiole segments, major pod vascular bundles or seed coats, but could be observed to some extent during photosynthate storage in pod wall pericarp tissues (Table IV). Little or no label was recovered from the cotyledonary insoluble (starch and protein) or lipid fractions.

Between 1 and 2.5 h after arrival of the ^{14}C-pulse in the fruit, 60% of the total radioactivity had passed into the cotyledons, with only 26 and 7% remaining in the seed coats and major pod veins, respectively (Fig. 3). By 2.5 and 5.5 h, label in the lipid fraction had increased to 6.7 and 8.9%, respectively. Only slight changes in ^{14}C distribution were observed between 2.5 and 5.5 h after arrival of ^{14}C-photosynthate in the fruit.

DISCUSSION

The principle sink for photosynthate in reproductive soybeans is the fruit pod and seeds (15). Translocation of ^{14}C-photosynthate during rapid pod filling is primarily to pods at the axil of the fed leaf, with significant levels of radioactivity imported by 1 h after feeding (2). Published data, however, provide no direct evidence of rates of uptake by the seeds or information regarding the roles played by various fruit tissues in photosynthate unloading and metabolism.

Under these experimental conditions, the [^{14}C]sucrose translocation velocity was estimated to be 0.9 cm min^{-1}, similar to the 0.78 cm min^{-1} previously reported for soybeans (13). Label could be recovered from fruit within 30 min of exposing the leaf at that node to $^{14}CO_2$ (Fig. 1). As the [^{14}C]sucrose entered and transversed the pods in three major vascular bundles, it was quickly partitioned between secondary bundles supplying the pod wall pericarp and the seeds. Vascularization of the pod wall is extensive, but only a single secondary bundle enters each seed borne alternately along the two ventral bundles of the pod (Fig. 2). Despite this, the seeds were the overwhelming sink for entering [^{14}C]sucrose, becoming radioactive immediately as compared to a 30-min lag before label appeared in the pod walls (Table I). Since the pod walls are known to recycle photosynthetically respired CO_2 to the developing seeds (21), a high photosynthate level in the minor veins of the pericarp might effectively reduce partitioning to the pod wall except in those cultivars which temporarily accumulate the recycled carbon (25). Translocation of [^{14}C]sucrose from the pod ventral bundles to the developing soybean cotyledons of the seed occurred via the seed coat (Fig. 3). Sucrose was rapidly unloaded from the highly branched vascular system of the seed coat; label accumulated almost linearly for 60 min to a level 3-fold greater than the supplying pod veins (Fig. 4).

The accumulation of entering ^{14}C-photosynthate by the seed coat during the lag in uptake by the cotyledons (Fig. 3) also exposes a mechanistic requirement for continued seed import. Mass flow of sucrose in soybean plants is assumed to occur down a concentration gradient from the leaf to areas of utilization, or sinks (6, 13) with the active loading of photosynthate in leaf minor veins generating the turgor pressure necessary to drive translocation (7). Unloading at the other end of the system produces a region of reduced osmotic pressure, a phloem concentration gradient, and maintains import (6, 7, 13). Continued rapid import is facilitated by sucrose hydrolysis, conversion to insoluble products, or compartmentation (8, 26). Since conversion of seed coat [^{14}C]-sucrose to other radioactive products did not occur, compartmentation of unloaded sucrose is indicated. The apoplast of the cell layers between the seed coat phloem and the inner epidermal surface (endothelium) is the most probable location. Support for this conclusion was given by (a) the lag between accumulation of label in the seed coats and passage into the cotyledons, (b) the lack of restriction of ^{14}C to the venation of the seed coats as determined by autoradiography, and (c) continued, rapid [^{14}C]-sucrose import and unloading into a tissue with an extractable sucrose pool of 7.4% of the seed coat dry weight (Table II). The

Table IV. *Distribution of ^{14}C among Photosynthate Fractions in Various Fruit Tissues 60 min after Arrival of ^{14}C-Photosynthate in Soybean Fruit*

Each value is the mean distribution ± SE of appropriate tissues of three fruit labeled and harvested as described in the text. Average radioactivity in fruit = 8.7×10^4 dpm.

	^{14}C Distribution Among Fractions				
Fruit Tissue	Neutral Sugars	Amino Acids	Organic Acids	Insoluble	Radioactivity in This Fruit
		%			*% of total*
Pod veins	92.6 ± 0.6	3.0 ± 0.1	3.5 ± 0.5	1.0 ± 0.6	20.4 ± 1.6
Seed coats	93.6 ± 0.4	3.2 ± 0.5	3.0 ± 0.1	0.2 ± 0.03	58.7 ± 3.3
Cotyledons	73.7 ± 3.1	12.4 ± 1.3	12.3 ± 2.1	1.6 ± 0.8	7.4 ± 1.7
Pod walls	87.6 ± 1.0	4.1 ± 0.2	7.2 ± 1.9	1.1 ± 0.7	13.5 ± 1.2
					100

actual sucrose concentration in the unloading zone is probably much greater, for several cell layers near the outer surface of the seed coat can be eliminated on a structural basis as sites for sucrose accumulation, and autoradiography indicated that the label was near the inner surface of the seed coat.

CONCLUSIONS

The rapid unloading and temporary accumulation of arriving sucrose in a compartment within the seed coat appears to be primarily responsible for the overwhelming partitioning of photosynthate to developing soybean seeds. Accumulation of sucrose in such a compartment, away from the phloem, would effectively maintain a steep concentration gradient from the leaf and import, at least in the short-term. Continued uptake and utilization by the cotyledons, is necessary for continued partitioning to the fruit. Accumulation of [^{14}C]sucrose in the seed coat probably occurs in the apoplast of the innermost cell layers, where it then diffuses away from the vascular bundles toward the inner epidermis (endothelium) of the seed coat prior to uptake by the cotyledons. Label then slowly enters the cotyledons from the surrounding seed coat (Fig. 3); there is no vascular connection between the maternal seed coat and the enclosed developing embryo. The cotyledons appear to absorb intact the [^{14}C]sucrose released from the endothelium of the seed coat, with hydrolysis and conversion to other products soon after uptake (Table III). This is consistent with studies of sucrose uptake by cotyledons of germinating *Ricinus* seeds (17), and observations of sucrose-dependent membrane depolarization in soybean cotyledonary cells (18), possibly suggesting an uptake mechanism in the cotyledons similar to that observed during loading of sucrose in minor leaf veins (9). Little is presently known about this transfer of photosynthate but release by the seed coat and absorption by the cotyledons may be aided by specialized transfer cells in either tissue (10).

Acknowledgment—The technical assistance of J. A. Louys during part of this study was sincerely appreciated.

LITERATURE CITED

1. Bils RF, RW Howell 1963 Biochemical and cytological changes in developing soybean cotyledons. Crop Sci 3: 304–308
2. Blomquist RV, CA Kust 1971 Translocation pattern of soybeans as affected by growth substances and maturity. Crop Sci 11: 390–392
3. Carlson JB 1975 Morphology. *In* BE Caldwell, ed, Soybeans: Improvement, Production, and Uses. Am Soc Agron, Madison, WI pp 17–95
4. Egli DB 1975 Rate of accumulation of dry weight in seed of soybeans and its relationship to yield. Can J Plant Sci 55: 215–219
5. Fisher DB 1970 Kinetics of C-14 translocation in soybean. I. Kinetics in the stem. Plant Physiol 45: 107–113
6. Fisher DB 1978 An evaluation of the Munch hypothesis for phloem transport in soybean. Planta 139: 25–28
7. Geiger DR 1975 Phloem loading. *In* MH Zimmermann, JA Milburn, eds, Transport in Plants I. Phloem Transport. Encyclopedia of Plant Physiology. Springer-Verlag, Berlin, pp 395–431
8. Giaquinta RT 1978 Source and sink leaf metabolism in relation to phloem translocation. Plant Physiol 61: 380–385
9. Giaquinta RT 1979 Phloem loading of sucrose. Involvement of membrane ATPase and proton transport. Plant Physiol 63: 744–748
10. Gunning BES, JS Pate 1974 Transfer cells. *In* AW Robards, ed, Dynamic Aspects of Plant Ultrastructure. McGraw-Hill, London, pp 441–480
11. Hardham AR 1976 Structural aspects of the pathways of nutrient flow to the developing embryo and cotyledons of *Pisum sativum* L. Aust J Bot 24: 711–721
12. Hardman LL, WA Brun 1971 Effect of atmospheric carbon dioxide enrichment at different developmental stages on growth and yield components of soybeans. Crop Sci 11: 886–888
13. Housley TL, DB Fisher 1977 Estimation of osmotic gradients in soybean sieve tubes. Qualified support for the Munch hypothesis. Plant Physiol 59: 701–706
14. Howell RW 1962 What happens to the energy of soybeans and corn? Better Crops with Plant Food 46: 4–5
15. Hume DJ, JG Criswell 1973 Distribution and utilization of ^{14}C-labelled assimilates in soybeans. Crop Sci 13: 519–524
16. Jenner CF 1974 An investigation of the association between the hydrolysis of sucrose and its absorption by grains of wheat. Aust J Plant Physiol 1: 319–329
17. Komor E 1977 Sucrose uptake by cotyledons of *Ricinus communis* L.: characteristics, mechanism, and regulation. Planta 137: 119–131
18. Litchner FT, RM Spanswick 1979 Sucrose co-transport in developing soybean cotyledons. Plant Physiol 63: S-341
19. Noble RD, DW Long, JW Burley 1973 Photosynthetic ^{14}C-labelling patterns in developing soybeans. Can J Bot 51: 1505–1512
20. Pamplin R 1963 The anatomical development of the ovule and seed in the soybean. PhD thesis. University of Illinois. Urbana
21. Quebedeaux B, R Chollet 1975 Growth and development of the soybean (*Glycine max* [L.] Merr.) pods. CO_2 exchange and enzyme studies. Plant Physiol 55: 745–748
22. Sakri FAK, JC Shannon 1975 Movement of ^{14}C-labeled sugars into kernels of wheat (*Triticum aestivum* L.) Plant Physiol 55: 881–889
23. Servaites JC, LE Schrader, DM Jung 1979 Energy-dependent loading of amino acids and sucrose into the phloem of soybean. Plant Physiol 64: 546–550
24. Streeter JG, DL Jeffers 1979 Distribution of total nonstructural carbohydrates in soybean plants having increased reproductive load. Crop Sci 19: 729–734
25. Thorne, JH 1979 Redistribution of assimilates from soybean pod walls during seed development. Agron J 71: 812–816
26. Wyse R 1979 Sucrose uptake by sugar beet tap root tissue. Plant Physiol 64: 837–841
27. Zee SY, TP O'Brien 1970 A special type of tracheary element associated with "xylem discontinuity" in the floral axis of wheat. Aust J Biol Sci 23: 783–791