Carbon-14 Distribution in Peanut Fruit Parts During Maturation Following ^{14}CO Treatment of Intact Plants[1]

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ABSTRACT

Effects of sampling date and developmental stage on the distribution of radioactivity within the crude ethanol, lipid, and starch fractions from fruit, seed coat, and seed of peanut were investigated. Major differences were found between the first and fourth feeding dates in the amount of ^{14}C-labeled photosynthate translocated to individual peanut fruit parts. Maximum levels of radioactivity in the pericarp, seed coat, and seed were attained at progressively later developmental stages as the respective part became the dominant metabolic sink. Within the fruit, maximum radioactivity in starch was reached during early maturity (stage 3) and total radioactivity generally decreased with successive feeding dates. Thus the level of photosynthate being translocated to a given fruit decreases as more fruit develop on the plant. Observed relationships between level of radioactivity and specific activity of fruit-part components were interpreted as indicating that metabolic reserves are built up in the fruit and seed coat during early maturation and utilized later during seed development and maturation when the level of available translocated photosynthate has diminished.

Translocation of labelled photosynthate from leaves to various plant parts has been the subject of several studies and reviews (Wardlaw, 1968; Milthorpe and Moorby, 1969). Distribution of the label has been determined in carbohydrates (Shannon, 1968, and 1972; Shannon and Dougherty, 1972) and in lipids (Beringer, 1971). Using autoradiographic techniques Khan and Akosu (1971) studied distribution of labelled photosynthates in peanut vegetative portions and fruits as influenced by plant development and position of the treated leaf. Apices, young expanding leaves, and roots were the major sinks until time of peg formation when pods became the major sinks.

Studies by Pickett (1950), Schenk (1961) and Pattee and coworkers (1974) have provided information on changes in chemical composition of the peanut seed and other fruit parts during maturation but little if any information is available concerning the effect of maturation and date of feeding on distribution of labelled photosynthates in peanut fruit parts. It was the objective of this study to provide such information.

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Materials and Methods

On July 12, August 3, August 29, and September 19, 1971, twelve peanut plants, variety NC-2, were randomly selected in a peanut field at the Central Crops Research Station, Clayton, North Carolina and exposed to $^{14}CO_2$. Each plant was enclosed with a $^{14}CO_2$ generating and exposure system (Mohapatra and Pattee, 1973) which contained 70 mCi of $Na_2\ ^{14}CO_3$. The $^{14}CO_2$ was released by addition of 2 ml of concentrated lactic acid and was circulated through the system by means of a hand pump, initially for 5 min. and then for one min. at approximately 15-min. intervals until 1.5 hrs. had elapsed. The generating and exposure system was then removed and the plants left in the field for 24 hrs. before being harvested, packed in ice and transported to the laboratory. All feedings commenced at 9 A.M. and no apparent differences were observed in the climatic conditions of the selected feeding dates.

The fruit were washed with tap water to remove soil, opened, and classified into maturity stages according to critieria described by Pattee, **et al.** (1974). The fruit was not segregated into individual parts prior to development stage 4. Commencing at development stage 4 the fruit were divided into pericarp, seed coat, and seed and fruit parts composited into samples large enough for analysis. These fruit parts have been defined and the extraction, separation, and analytical procedures for the "crude ethanol-soluble" (EtOH), starch, and lipid fractions described by Pattee, **et al.** (1974). Radioactivity measurements were made with a Packard liquid scintillation spectrometer with external standardization according to the method of Hayes (1963).

Results and Discussion

Pattee and coworkers (1974) have suggested that the role of regulating substrate supply to the developing seed shifts from the pericarp to the seed coat with ontogeny of the complete peanut fruit. This suggestion is supported by the distribution of radioactivity in the crude EtOH fractions (Figure 1). These data also confirm the observations by Khan and Akosu (1971) that soon after peg formation, the complete peanut fruit becomes a sink for photosynthate produced by the leaves. Generally values for each component were low at the youngest developmental stage, increased to a maximum in subsequent developmental stages, and then decreased. The fruit/pericarp (Figure 1A) reached maximum radioactivity levels at stage 3 for the first two feeding dates and not until stage 5 at the third feeding date. At the fourth feeding date, the pericarp appears to be no longer a major metabolic sink and metabolism of ^{14}C labelled photosynthate decreased with increased maturity; however, the increased number of fruit to which labelled photosynthate must be distributed is a factor which can not be completely evaluated (Table 1). These data do suggest that the sampling period selected during the growing season for $^{14}CO_2$ feeding has an

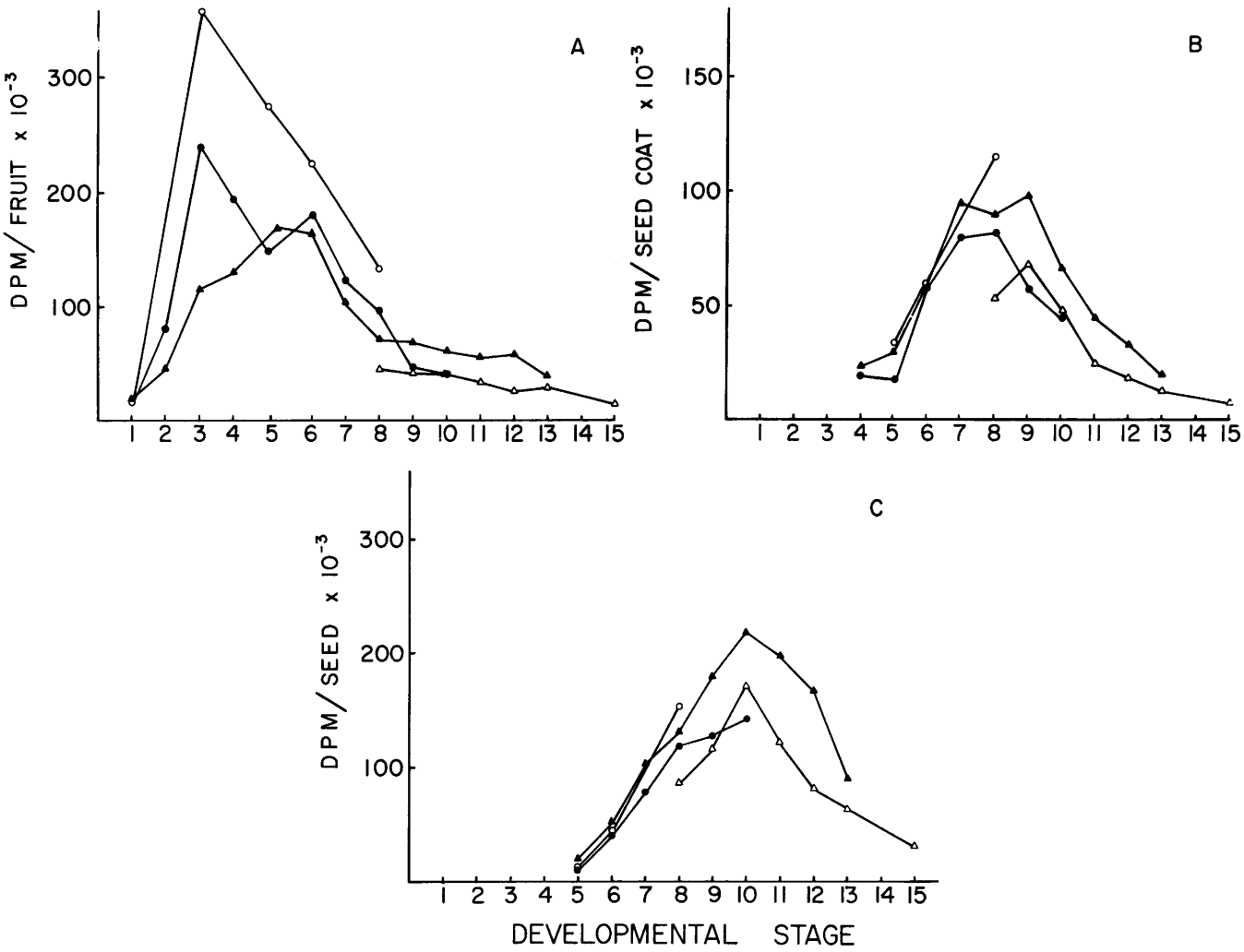

Fig. 1 Changes in the radioactivity level of the crude ethanol-soluble fraction from peanut fruit: beyond stage 3 - pericarp (A), seed coat (B) and seed (C) as influenced by developmental stage at: First feeding (0 - 0); Second feeding (● - ●); Third feeding (△ - △); Fourth feeding (▲ - ▲).

influence on the results obtained for the peanut fruit.

Seed coats contained maximum radioactivity levels in the crude EtOH-soluble fraction between developmental stage 7 and 9 (Figure 1B). In contrast to the fruit, the sampling period during the growing season selected for $^{14}CO_2$ exposure appeared to have little if any effect on the distribution pattern observed. Comparison of the radioactivity in the crude EtOH-soluble fraction from the seed coat with the sugar content reported by Pattee and coworkers (1974) indicates similar trends. Such similarity might be expected since sugars account for up to 90% of the radioactivity being translocated to the peanut fruit (Mohapatra and Pattee, 1973).

Between stages 9 and 10 major radioactive accumulations of photosynthate shifted from seed coat to seed with maximum seed radioactivity occurring at stage 10 (Figure 1C). A maximum at this stage might not be expected since sugar content in peanut seeds has been reported to continue increasing throughout maturation (Pattee *et al.*, 1974). Such results suggest that the sugars accumulated in the later stages of seed maturation are coming from a non-radioactive reserve source rather than from radioactive translocated photosynthate; thus, the diminished incorporation of radioactivity into the sugars at the later stages of development.

Radioactivity in the lipid fraction of the seed (Figure 2A) increased rapidly from the initial level at stage 5 to a maximum at stage 9 and then decreased rapidly until at stage 13 and beyond little accumulation was observed Comparison of the level of radioactivity in the lipid fraction obtained at stage 8 in the first and fourth feeding dates suggest that major changes are occurring in the specific activity of the photosynthate being translocated from the aerial portion of the plant. Specific activity levels of the lipid also support this suggestion (Figure 2B). Lipid specific activity values for comparable maturation stages were higher on the first feeding date than on subsequent feeding dates. However the specific activity data from the fourth feeding date indicate that the low radioactivity level per seed was due to the increased number of fruit on the plant to

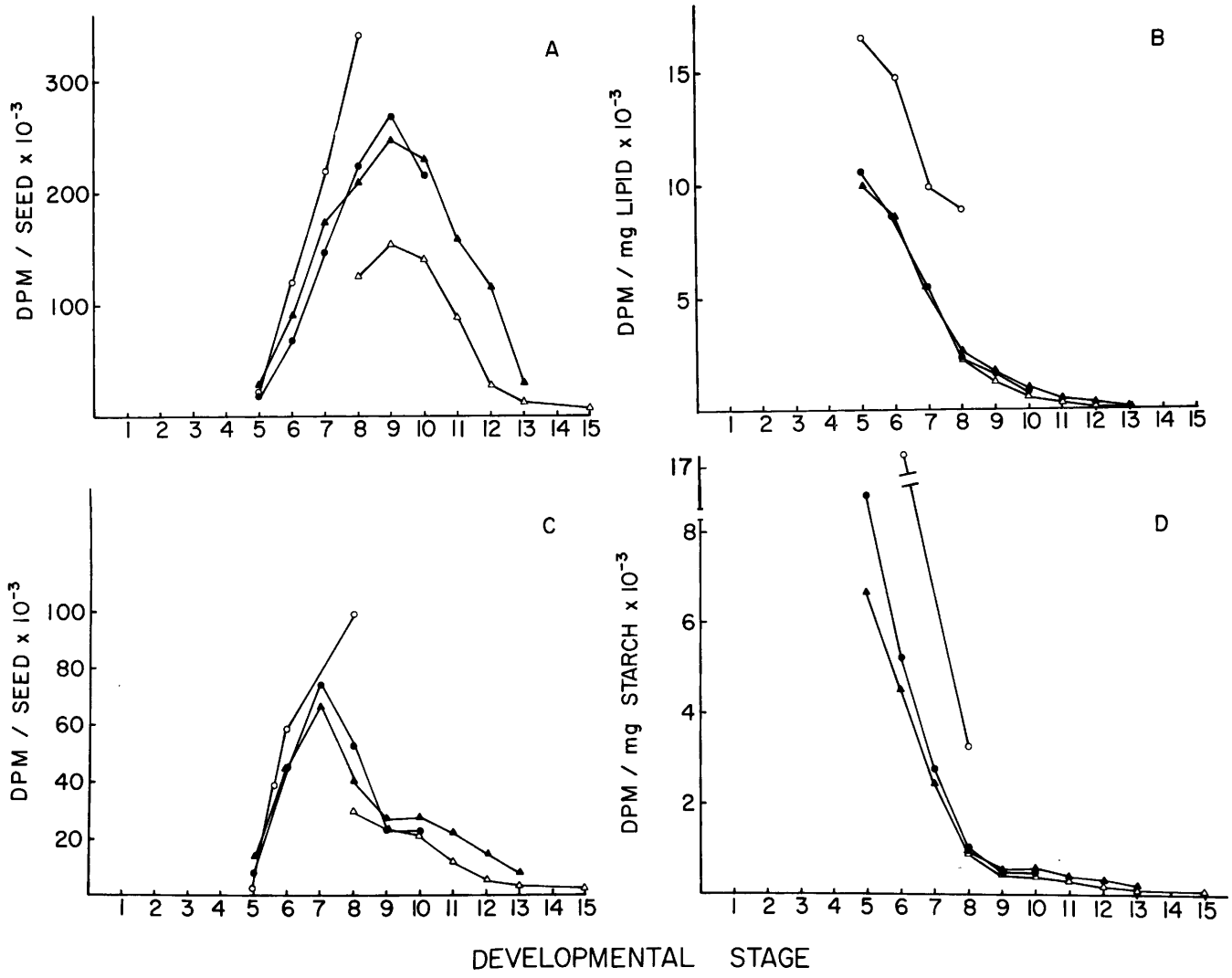

Fig. 2. Changes in the radioactivity of peanut seed components: Lipids (A) and (B); Starch (C) and (D) as influenced by developmental stage at selected feeding dates. Symbol legend same as Figure 1.

which the labelled photosynthate must be distributed since specific activity values were comparable for the second, third and fourth feeding dates.

Changes in fresh weight, dry weight, and number of seeds per plant are given in Table 1. These data show the growth pattern of the aerial portion of the plant as well as the increase in number of seeds which must be supported by the aerial portion. Number of seeds per plant increased almost 3-fold between the first and fourth feeding dates. Such an increase would have considerable effect on the distribution of radioactivity on a "per seed" basis given a constant initially available amount of radioactivity. Changes in the distribution of developmental stages present at a given feeding date can be seen in Figure 1. The first feeding date had developmental stages ranging from 1 through 8 while the second and third feeding dates ranged from 1 through 10 and 13, respectively. The developmental stages present at the fourth feeding date, in adequate quantities to analyze, ranged from stage 8 to stage 15. Thus, due to the indeterminate flowering habit of peanut plants a wide range of maturity stages were found on each plant and with later feeding dates the average range shifted from an immature grouping to a mature grouping. All peanuts analyzed from the fourth feeding date would have been of marketable size since Pattee (unpublished data) has found that developmental stage 7 kernels will ride a 15/64 X 1 inch slotted screen.

Table 1. Observations on plant fresh weight, plant dry weight and fruit number per plant during the 1971 growing season.

Date	Av. Plant Fresh Wt. (gm)	Av. Plant Dry Wt. (gm)	Number of fruits per plant
July 12	76[a]	30	18
August 3	213	42	34
August 29	167	36	31
September 19	310	71	53

[a]All values the average of four replications. Three plants per replication.

Radioactivity levels in the starch fraction (Figure 2C) of the seed do not appear to be as subject to feeding date variations as the lipid fraction. The maximum radioactivity level occurred at developmental stage 7 then decreased rapidly to stage 9 and gradually decreased beyond stage 9. Specific activity of the starch fraction (Figure 2D) was also higher in the first feeding than in the other feedings. The sharp decrease in specific activity through developmental stage 8 suggests that rapid starch synthesis was occurring up to this stage of seed development. This agrees with observations by Pattee and coworkers (1974) which showed maximum starch levels in the seed at approximately stage 9.

Changes in the starch radioactivity levels in the pericarp and seed coat are shown in Figure 3. The starch fraction in the pericarp (Figure 3A) reached a maximum radioactivity level at stage 3 in the first and second feeding dates and but not until stage 5 in the third feeding date. Very little radioactivity was found in the starch fraction from the fourth feeding date. The generally lower radioactivity levels observed in successive feeding dates may suggest that seed developing late in the growing season receive less photosynthate per unit of time than early-developing seed because the total available photosynthate is divided among a larger number of seeds. Since starch accumulation appears to have occurred during all feeding periods where early developmental stages were available, the level of photosynthate being translocated to the fruit was probably not a limiting growth factor during the 1971 growing season. It is possible that the level of photosynthate being translocated to the fruit acts as a growth-regulating mechanism and low levels of photosynthate might induce the initiation of maturation processes in the seed. Such a condition might explain the early maturation of peanut crops during growing seasons with limiting moisture levels.

The specific activity of the fruit starch (Figure 3B) suggests major accumulation of starch was taking place during the first feeding date. The pat-

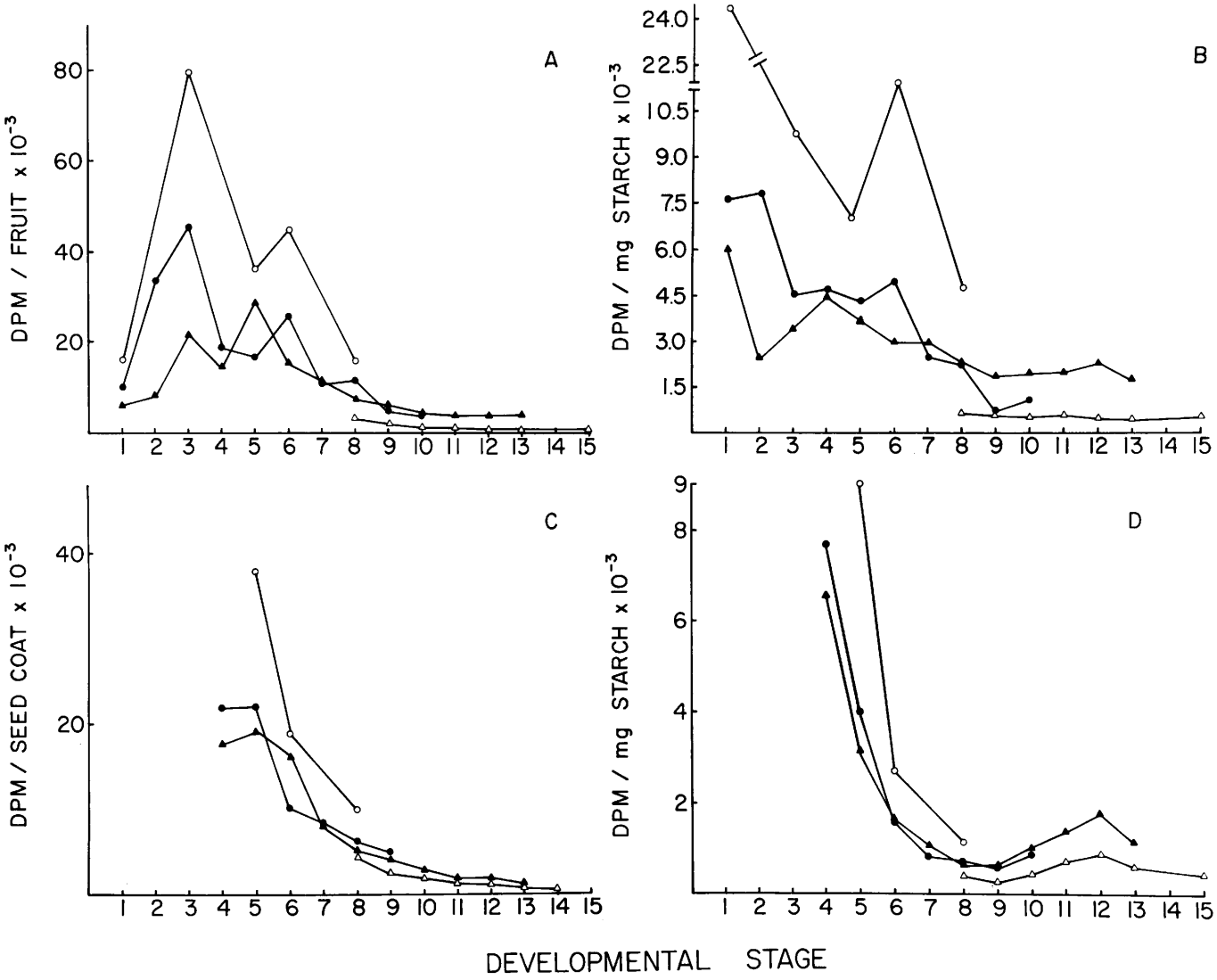

Fig. 3. Changes in the radioactivity of the starch fraction from peanut fruit: beyond stage 3 - preicarp (A) and (B); seed coat (C) and (D) as influenced by developmental stage at selected feeding dates. Symbol legend same as Figure 1.

tern of specific activity in the third feeding date suggests that synthesis and utilization were taking place simultaneously since the specific activity values for starch did not decrease to as low a level as other constituents examined.

Radioactivity levels of the starch fraction from the seed coats were at or near maximum in developmental stages 4, 5, and 6 for all feeding dates (Figure 3C). The decline in radioactivity with development suggests that the rate of photosynthate incorporation into seed coat starch declines with maturity. However, starch specific activity (Figure 3D) indicates that until approximately stage 7 or 8 accumulations of starch were occurring in the seed coat. The increase in specific activity in the later developmental stages indicates that some starch synthesis must be occurring in the seed coat throughout development and maturation (Figure 3C).

The data presented in this and the companion study by Pattee *et al.* (1974) have pointed out the changing metabolic role of fruit and seed coat in supporting the development of the seed. The main photosynthate translocated in peanuts is sucrose (Mohapatra and Pattee, 1973) and up to developmental stage 6 the main extractable reserve repository of this translocate is starch. Beyond this stage of development the formation of lipid reserve becomes the main metabolic sink for photosynthetic material.

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