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ENHANCED RATE OF ^{14}C-SOLUTE RELEASE TO THE FREE SPACE BY THE PHLOEM OF VICIA FABA STEMS PARASITISED BY CUSCUTA

P. WOLSWINKEL

Botanisch Laboratorium, Utrecht

SUMMARY

By washing out ^{14}C-labelled assimilates from the free space of stem segments of *Vicia faba* L., considerably more label could be removed from a stem parasitised by *Cuscuta* species than from a normal one. This could mean that the moving of assimilates from the host phloem to the parasite at least partly occurs through the apoplast.

The assumed unloading of the host phloem is apparently under metabolic control since it appeared to be inhibited at 0 °C and after addition of dinitrophenol.

1. INTRODUCTION

The stem parasite *Cuscuta* usually has a very deleterious influence, draining the phloem of the host. Using ^{14}C-labelled metabolites it could be shown in a previous study (WOLSWINKEL 1974) that *Cuscuta* is able to withdraw almost all assimilates which normally move from a photosynthesizing host leaf to growing pods and seeds.

A great enlargement of the surface area of the absorptive parts of the haustorial organ (SCHUMACHER 1934, DÖRR 1967, 1968a, c and 1972) emerges as a feature seeming to be essential for the intensive transfer of metabolites. The walls of the hyphae growing in the direction of the vascular bundles ("Suchhyphen") are perforated by numerous plasmodesmata ("Aussenwandplasmodesmen") which, however, are lacking at the contact sites with the sieve tubes SCHUMACHER & HALBSGUTH 1939, DÖRR 1967, 1968b and 1969, KOLLMANN & DÖRR 1969). Since Schumacher and Halbsguth could not find plasmodesmata in the parts of an absorptive hypha adjacent to the sieve tube of the host ("Anschlusshyphe"), they took for proved that the intensive transfer of solutes from the host sieve tube to the parasite has to occur freely through both cell walls, without mediation of plasmodesmata. They stressed the fact that the parasite never penetrates into the lumen of the sieve tube but nevertheless has the capacity of taking solutes out of it. This seems an indication that this kind of solute withdrawal in some way resembles the normal moving out of solutes from a sieve tube into the surrounding tissues. KOLLMANN & DÖRR (1969) have postulated that all solutes first have to leave the sieve element through its outer membrane and pass on to the free space of the cell walls before they finally are absorbed by the plasmalemma of the parasitic cell.

GUNNING & PATE (1969) and PATE & GUNNING (1972) have presented review papers on "transfer cells", plant cells with wall ingrowths, apparently specialised in relation to short distance transport of solutes. Numerous mitochondria and a conspicuous endoplasmic reticulum usually accompany the wall specialization. The plasma membrane follows the contours of the wall and cytoplasm penetrates between the individual ingrowths. Such transfer cells are restricted to situations where adverse surface area-volume relationships exist between donor and receptor compartments of the transport pathway and/or where the transported solutes are accompanied by a minimal flow of solvent. In the case of the absorptive hyphae of *Cuscuta* the function of the transfer cell can be related to absorption of solutes from an extracytoplasmic compartment.

Using the method of washing out sugars from the free space of isolated parts of leaves of sugar beet, KURSANOV & BROVCHENKO (1969, 1970) found a characteristic difference between mesophyll tissues and conducting bundle tissues. When sugars are removed from the free space of the mesophyll, it is refilled with sugars from the inner space of the cells. If conducting bundles are leached, the free space is not filled again. The data of Kursanov and Brovchenko seem to indicate that assimilates, at least at a certain stage of their movement, are released from the symplast of photosynthesising cells, enter the apoplast and subsequently are reabsorbed from the free space by phloem cells. Also according to GEIGER et al. (1971), who reviewed structural evidence in the light of the physiological observations of Kursanov and Brovchenko, in sugar beet leaves the sugar appears to enter the apoplast prior to vein loading.

According to JACOB & NEUMANN (1968) the phloem of the host is rendered leaky by the presence of *Cuscuta*. This is a plausible idea, found elsewhere in the literature on *Cuscuta* (e.g. SCHUMACHER & HALBSGUTH 1939), but the present author is not aware of any report in the literature of experimental data on this problem.

Vicia faba, with its hollow stem, seemed to us a very suitable host plant to approach this question. It may be assumed that in general, a few hours after administration of label to a leaf, the major part of the ^{14}C-solutes which have arrived in the stem will be present in the vascular bundles. As far as the ^{14}C-solutes are concerned stem segments of broad bean seem comparable with the tissues of fine conducting bundles of leaves prepared by Kursanov and Brovchenko. In experiments on washing out solutes from the free space by free diffusion very small pieces of tissue are normally used in order to remove all the solutes from the free space (e.g. GLASCIOU & GAYLER 1972). From the thin tissue cylinder of broad bean stem segments solutes present in the free space can be washed out with water. In a similar way a part of a host stem, to which a haustorial coil is attached, can be used. The present paper describes such washing experiments and shows a stimulated unloading of host stem tissues by the presence of the parasite haustoria.

Fig. 1 shows a stem segment with haustorial coil as used in the experiments and *fig. 2* presents a scheme of the anatomical situation of the place where the process studied in this paper is supposed to occur.

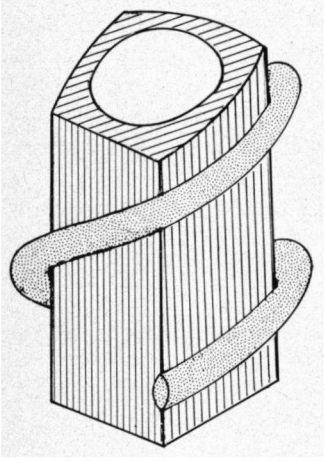

Fig. 1. Scheme of hollow stem segment of *Vicia faba* with haustorial coil.

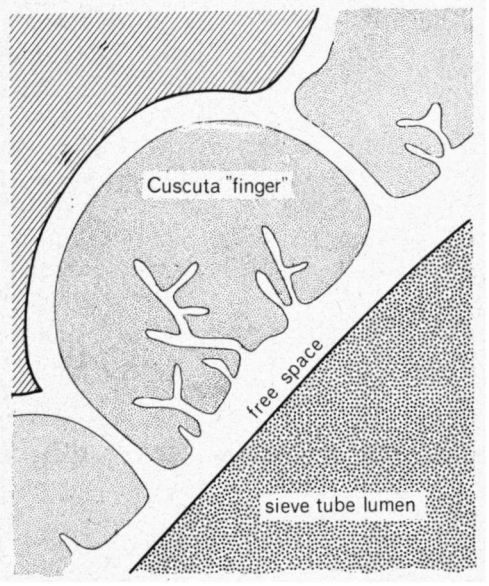

Fig. 2. Scheme of anatomical situation in the contact region of host and parasite. The *Cuscuta* "fingers" are situated between sieve tubes and other phloem cells of the host and show wall ingrowths on the side directed towards the sieve tube (according to data of Dörr).

2. MATERIAL AND METHODS

2.1. Cultivation of plants

Host and parasite were grown as described in a previous paper (WOLSWINKEL 1974). *Vicia faba* L. cv. Witkiem was used as host and four species of the genus *Cuscuta* were used as parasite, viz. *C. campestris* Yunck., *C. europaea* L., *C. lupuliformis* Krocker, and *C. reflexa* Roxb. Seeds of broad beans of *tables 1* and *2* were planted in the garden on 27-4-1972 and those of the hosts of *tables 3* and *4* on 22-8-1972. The plants of *tables 5, 6* and *7* were grown on a nutrient solution in the greenhouse and used as young plants.

2.2. Application of $^{14}CO_2$, ^{14}C-sucrose and ^{14}C-L-alanine to Vicia faba

$^{14}CO_2$ was administered to a single leaf as described in a previous paper (WOLSWINKEL 1974).

For the application of sucrose and amino acids two methods were used:

a. the sugar was administered through a severed petiole or main vein of the leaf. This was immersed in a narrow glass tube, usually containing a solution of 0.3 ml with 3 μCi of sucrose-^{14}C(U), 9.6 mCi/mmol which subsequently was

gradually sucked up by the xylem system in basal direction. The distribution pattern of the ^{14}C, introduced in this way into the plant indicates, however, a rapid entry into other tissues – e.g. the phloem – as well, in accordance with data from literature (e.g. STOUT & HOAGLAND 1939). In experiments with the amino acid usually also 3 μCi prepared from L-alanine-^{14}C(U), 10 mCi/mmol was used. b. the leaf flap method (BIDDULPH 1941) in which a ^{14}C-solute solution was applied to a leaflet. The midvein of the leaflet was severed a few cm from the apex and the apical part was subsequently brought into a narrow glass tube containing the ^{14}C-solution, the solution being sucked up in this way in apical direction; this application results in phloem movement out of the leaf.

All ^{14}C-chemicals were obtained from the Radiochemical Centre, Amersham, Geat Britain.

2.3. Analysis of ^{14}C-labelled plants

Stem segments and other parts of host and parasite were extracted and the ^{14}C-content counted as described in a previous paper (WOLSWINKEL 1974).

2.4. The procedure of washing out ^{14}C-solutes from the free space of hollow stem segments

After removing the filaments of *Cuscuta* from the haustorial coil by cutting, a stem part was divided into four successive segments (most times four successive internodes of about 3–4 cm length, nodes usually being excluded). Very long internodes were divided in two or three parts of about 3–4 cm and, when used as one sample, the different pieces were shaken in the same flask. In many experiments the wound surfaces on the haustorial coil were covered with silicone rubber (SIL 21 elastic impression paste was mixed with SIL 21 accelerator) to prevent interference during washing by ^{14}C-solutes originating from *Cuscuta* haustorial coil wounds, but the results did not differ clearly from those obtained in experiments without covering.

The stem segments with or without haustorial coils were transferred to Erlenmeyer flasks of 300 ml containing 50 ml of demineralized water and shaken 120 times per minute in a horizontal direction in a water bath at 25 °C. After 30 min. the water was decanted, another 50 ml water was added and washed for a second period and afterwards for a third period of 30 min. The washing solutions obtained were evaporated on a steam bath, the residue was dissolved in 0.5 ml water and counted. The sum of ^{14}C in the washing solutions was added to the quantity of ^{14}C remaining in the stem segments.

In the tables, column 3 gives the values obtained in this way, divided by the fresh weight (g) of the segments, as "initial ^{14}C-content" in cpm/g. The sign* before the internode number in column 1 indicates the presence of a haustorial coil.

2.5. Numbering of plant parts

In the tables, leaves and internodes are numbered. Leaf 1 and 2 are the small reduced leaves at the stem base and leaf 3 is the first normal leaf. The internode

number corresponds with that of the leaf above it. In the tables, the sign (u) after a numbered internode means the upper half of the internode is sampled, (l) means the lower half is sampled.

2.6. Presentation of results

In the tables, data of some typical experiments are presented. In spite of strong variations between the values of the percentage of ^{14}C washed out during the three washing periods (depending, among other things, on the vigour of the parasite, the stage of development of host and parasite, the growing conditions, the number of windings of the haustorial coil around the stem segment of the host, the stem part used, the species of *Cuscuta*, and the way of administering ^{14}C to the host), the phenomenon described was found clearly in most experiments. The experiments were repeated several times and the phenomenon appeared to be common in spite of quantitative variations.

3. RESULTS

Table 1 shows an experiment with a non-parasitised plant and with a host parasitised by *C. lupuliformis*. In the non-parasitised plant most ^{14}C is washed out during the first 30 min., values becoming higher for more basally placed stem segments. In the following two periods of 30 min. considerably less ^{14}C appears to be washed out. In the experiment with the parasitised host for the first period much higher values are found than those for the control plant. The same applies for the two following periods. Only the stem part consisting of the upper half of int. 12 + int. 13 + int. 14 + the lower half of int. 15 presents values comparable to control. This long part of the stem, which is not parasitised, behaves as a non-parasitised stem. The upper part of int. 11 + int. 10 seems to be influenced by the neighbourhood of *Cuscuta*.

Table 2 presents results of an experiment, later in the season, with a non-

Table 1. Redistribution of ^{14}C-sucrose administered via the petiole of leaf 13. After 4½ hrs. the stem is divided in segments. *: sites of attachment of *Cuscuta*.

internode number	weight (g)	initial total ^{14}C cpm/g × 10^2	% of ^{14}C washed out		
Non-parasitised plant (19-7-1972):			1st	2nd	3rd
15(u)+16	4.41	31.2	5.0 ± 0.5	1.6 ± 0.5	0.6 ± 0.2
12(u)+13+14+15(l)	5.80	313.9	8.7 ± 0.2	3.5 ± 0.1	1.4 ± 0.1
11(u)+12(l)	4.02	45.1	13.1 ± 0.5	1.1 ± 0.6	0.8 ± 0.3
10+11(l)	5.29	29.0	14.1 ± 0.6	5.5 ± 0.6	2.1 ± 0.4
Host parasitised by *C. lupuliformis* (18-7-1972):			1st	2nd	3rd
* 15(u)+16	4.27	40.0	38.2 ± 0.5	7.4 ± 0.3	4.7 ± 0.3
12(u)+13+14+15(l)	8.88	791.5	6.7 ± 0.1	1.6 ± 0.1	1.0 ± 0.1
* 11(u)+12(l)	3.84	103.7	26.4 ± 0.3	6.0 ± 0.2	4.3 ± 0.2
10+11(l)	4.50	31.5	37.7 ± 0.7	11.7 ± 0.4	7.4 ± 0.3

Table 2. Redistribution of ^{14}C-sucrose administered via the petiole of leaf 10 of a non-parasitised plant (10-8-1972). After 6 hrs. the stem is divided into segments.

internode number	weight (g)	initial total ^{14}C cpm/g × 10^2	% of ^{14}C washed out		
			1st	2nd	3rd
12	3.08	31.4	5.9 ± 1.3	1.1 ± 0.4	0.5 ± 0.3
11	3.57	59.5	8.3 ± 0.6	1.4 ± 0.3	0.9 ± 0.2
10	3.66	1370.9	6.9 ± 0.2	0.9 ± 0.1	0.4 ± 0.1
9	4.33	404.2	9.8 ± 0.3	2.1 ± 0.1	1.0 ± 0.1

parasitised plant with large pods of 20 cm; the plant was already senescing and several leaflets had already dropped off .Values for the first washing period are low and for the following ones very low, almost negligible. Parasitised stems in several experiments showed a behaviour comparable with the parasitised stem of *table 1.*

Table 3 shows the behaviour of a host parasitised by *C. europaea.* In this experiment the third period lasted 3 hours (instead of 30 min.) in order to study whether an important ^{14}C-release continues during the hours following the first 90 min. The values obtained for the sample consisting of the upper half of int. 6 + int. 7 are comparable with those of controls in may experiments. The parasitised stem segments show considerably higher values. Conspicuous also are the high levels for the second and third period of washing, which were obtained from parasitised stem segments and also, to a lesser degree, from int. 4, having a position between two segments of attachment.

In an experiment on 27-10-1972 comparable results were obtained with a set of plants in which $^{14}CO_2$ was administered to leaf 7. In the non-parasitised plant about 95% of the total quantity of ^{14}C washed out during the three periods was washed out during the first period and in the next two periods only a few % of that quantity were washed out. In the parasitised stem segments, however, in which a larger total quantity was washed out, about 40% of that quantity was washed out in the second and third period.

Table 3. Redistribution of ^{14}C-sucrose administered via a leaf flap (in this experiment, however, sucked up in basal direction) to leaf 6 of the host parasitised by *C. europaea.* After 5 hrs. the stem is divided into segments (23-10-1972).

internode number	weight (g)	initial total ^{14}C cpm/g × 10	% of ^{14}C washed out		
			1st	2nd	3rd
6(u)+7	8.39	35	11.4 ± 1.9	2.4 ± 1.0	2.1 ± 1.0
* 5+6(l)	8.88	119	20.4 ± 0.8	11.3 ± 0.4	18.0 ± 0.6
4	2.81	52	15.7 ± 3.9	10.0 ± 2.0	12.2 ± 1.7
* 3(u)	4.50	30	26.1 ± 3.8	14.8 ± 1.9	15.3 ± 1.9

Table 4. Translocation of ^{14}C-assimilates after application of $^{14}CO_2$ to leaf 7 of the host parasitised by *C. europaea*. After 10 hrs. the stem is divided into segments. Washing at 0 °C (1-11-1972).

internode number	weight (g)	initial total ^{14}C cpm/g × 10^2	% of ^{14}C washed out		
			1st	2nd	3rd
7	2.97	19.2	8.5 ± 1.0	3.6 ± 0.5	0.3 ± 0.2
* 6(u)	2.87	20.9	7.0 ± 0.9	2.3 ± 0.5	0.5 ± 0.2
* 5(u)+6(1)	3.08	27.1	8.7 ± 0.8	2.6 ± 0.4	0.4 ± 0.2
4+5(1)	3.18	8.1	14.4 ± 2.0	3.7 ± 1.2	0.3 ± 0.3

As shown in *table 4*, the effect of *Cuscuta* disappears when the stem segments are washed in water with a temperature of 0 °C. No higher values are found in the parasitised part of the stem. In this experiment at 0 °C the values of all four segments are much lower than those shown in *table 3*.

The experiments with broad bean plants grown in the garden, were afterwards repeated with young plants grown in the winter season in the greenhouse on a nutrient solution.

In the host of *table 5*, *C. reflexa* was first attached to the upper part of int. 3 and to the whole length of int. 4 and secondly to the greater part of int. 7. A strong stimulated filling of the free space with ^{14}C-solutes is shown in the parasitised segments. In all three periods the percentages obtained for parasitised segments are about twice as high as for the neighbouring non-parasitised segments.

In another experiment, in which *C. europaea* was attached to the top of int. 4 and the base of int. 5, sucrose was administered to leaf 7 and the stem segments were washed at 0 °C. Whereas for the two segments above the parasitised part values of 8.7 and 8.6 % were obtained in the first period of washing and a value of 7.3 % for the segment below the parasitised part, the parasitised part showed a value of 10.7 %. The values for the second and third period were very low and almost equal for all four segments. A larger quantity of ^{14}C-solutes seems to have been present in the free space of the parasitised stem part at the start of the washing procedure but a further filling of the free space was inhibited at 0 °C.

To investigate the cause of the inhibition of refilling of the free space at 0 °C

Table 5. Redistribution of ^{14}C-sucrose administered via the main vein of leaflet of leaf 6 of the host parasitised by *C. reflexa*. After 4½ hrs. the stem is divided into segments.

internode number	weight (g)	initial total ^{14}C cpm/g × 10^2	% of ^{14}C washed out		
			1st	2nd	3rd
* 7	0.61	32.2	19.4 ± 1.7	7.0 ± 0.9	3.1 ± 1.5
6	0.66	84.0	7.9 ± 0.7	3.7 ± 0.5	1.6 ± 0.6
5	0.83	65.5	12.4 ± 0.7	5.3 ± 0.6	3.5 ± 0.5
* 3(u)+4	1.19	52.8	20.9 ± 0.7	11.4 ± 0.6	6.1 ± 0.5

Table 6. Redistribution of ^{14}C-L-alanine administered via the main vein of a leaflet of leaf 7 of the host parasitised by *C. reflexa*. After 4 hrs. the stem is divided into segments.

internode number	weight (g)	initial total ^{14}C cpm/g $\times 10^2$	% of ^{14}C washed out		
			1st	2nd	3rd
7	0.43	95.1	5.4 ± 0.7	4.2 ± 0.7	3.1 ± 0.8
6	0.51	63.7	4.4 ± 0.8	5.6 ± 0.8	3.3 ± 0.7
*5	0.49	60.6	11.2 ± 1.1	6.3 ± 0.9	5.3 ± 1.2
4	0.60	35.1	7.3 ± 1.2	5.2 ± 1.2	2.2 ± 1.3

some further experiments were carried out with dinitrophenol, an uncoupler of oxidative phosphorylation. One could imagine that the structure of membranes is changed at 0 °C, preventing the normal moving out of assimilates from the phloem system. With 10^{-4}M dinitrophenol at room temperature, however, results were obtained comparable with the influence of a temperature of 0 °C. An active metabolism seems therefore a prerequisite for normal movement of assimilates out of the sieve tubes into the free space.

Table 6 shows a typical experiment on the behaviour of ^{14}C-L-alanine. This amino acid was used for studying the behaviour of amino acids because of its presence in important quantities in the free space of leaf blade tissues of sugar beet, suggesting a parallel with sugars (BROVCHENKO & RYABUSHKINA 1971), and its strong absorption by surrounding tissues during xylem translocation in tomato stems (VAN DIE & VONK 1967), suggesting stem tissues to be an important sink for L-alanine. As shown in *table 6*, in the parasitised stem segment an enhanced filling of the free space can be found in a similar way as in experiments in which ^{14}C-assimilates or ^{14}C-sucrose were used.

Washing in the presence of 10^{-4}M dinitrophenol also resulted in a reduction of the percentage of washed out label. As shown in *table 7*, only in the first period a very small influence of *Cuscuta* can be found. In the other two periods the values of the parasitised segment are comparable with those of the non-parasitised segments.

We can summarize our results by mentioning five phenomena:

1. Much higher percentages of ^{14}C are washed out from parasitised segments than from controls.

Table 7. Redistribution of ^{14}C-L-alanine administered via the petiole of leaf 7 of the host parasitised by *C. europaea*. After 6 hrs. the stem is divided into segments. Washed in 10^{-4}M dinitrophenol.

internode number	weight (g)	initial total ^{14}C cpm/g $\times 10^3$	% of ^{14}C washed out		
			1st	2nd	3rd
7	0.39	709.5	11.2 ± 0.1	1.5 ± 0.1	1.5 ± 0.1
6(u)	0.62	189.7	11.2 ± 0.1	2.7 ± 0.1	1.1 ± 0.1
*6(l)	0.23	206.9	13.6 ± 0.2	2.5 ± 0.1	1.8 ± 0.1
5	0.42	35.1	9.7 ± 0.3	1.3 ± 0.2	1.7 ± 0.2

2. A large portion of the ^{14}C washed out during three periods of 30 min. is washed out in the second and third period in parasitised segments; in non-parasitised segments this portion is very low.
3. The phenomena described under 1. and 2. do not occur if the temperature of the washing water is 0 °C.
4. By washing at 25 °C in 10^{-4}M dinitrophenol, the results obtained are comparable with those obtained by washing at 0 °C.
5. The behaviour of ^{14}C-solutes applied as amino acid is comparable with that of ^{14}C-sucrose and ^{14}C-assimilates.

We can add to this summary a remark concerning the use of four different *Cuscuta* species. All species show essentially the same influence, in spite of differences in their way of growing. *C. europaea* seems to be the species showing most clearly phenomenon 2.

4. DISCUSSION

Our experiments with parasitised stems indicate a change in permeability of the vascular tissues of the stem, normally engaged in longitudinal translocation of assimilates from leaves to sinks like the root or growing fruits. In plants not parasitised by *Cuscuta* relatively small quantities of substances are released to stem tissues surrounding the sieve tubes. A considerable part of the ^{14}C-solutes washed out from non-parasitised stems in the first washing period, will consist of solutes already in the free space of the stem before washing was started since the values of the first washing period at 0 °C are considerable although lower than those obtained at 25 °C. Only a small quantity of ^{14}C can be washed out during following periods, presumably for a part originating from incomplete washing during the first period. Kursanov and Brovchenko have obtained comparable results with vascular bundles from leaves. The high values for the last two washing periods, obtained on parasitised stem segments or segments bordering them, indicate a continuous movement of ^{14}C-solutes from the conducting tissue into the free space of the stem. In this respect, parasitised sieve tubes behave like mesophyll tissues of sugar beet, continuously refilling the free space with solutes.

To some extent, the situation is comparable with that in sugar beet roots in which unloading phloem supplies sugars to the parenchyma of storage tissues via the free space (KHOLODOVA et al. 1968, ENGEL et al. 1968). In parasitised stems, however, we find the special situation that a system specialised in long distance translocation of assimilates is drained by another one, whereas in leaves or sinks like sugar beet root, sugar cane stem and fruits vascular bundles border upon tissues typically consisting of parenchyma cells.

By the washing procedure a considerable part of the ^{14}C-solutes are withdrawn from the pool from which the transfer cells of the *Cuscuta*-haustorium normally absorb them. Because withdrawal by washing will not be complete owing to the result of the probable simultaneous absorption by *Cuscuta*, the amounts of ^{14}C-solute entering the free space may be higher than the measured

values indicate. Moreover, following sampling it is impossible to remove completely all the haustorial cells of *Cuscuta* from the bean stem segments. Remnants of the extensively radiating haustoria containing relatively much label will stay behind in the stem tissue. Too high values for ^{14}C remaining in washed out segments of the bean stem, result in too low values for the percentage of ^{14}C-solutes in the washing solutions. Both reasons, absorption by *Cuscuta* and the presence of haustorial cells in the extracted broad bean stems, cause the real differences in the quantity of ^{14}C-solutes, released to the free space to be more pronounced than the values measured.

A parasitised stem part represents a very strong sink. One could imagine that an enhanced removal of assimilates from the free space by *Cuscuta* would result in a shift in the balance between loading and unloading of the phloem in the direction of a more important unloading. By washing, however, solutes in the free space can also be withdrawn from the pool from which cells bordering the phloem normally absorb them, but non-parasitised stems cannot be stimulated to a similar enhanced unloading in this way.

The fact that non-parasitised stem segments situated near parasitised parts, can also be influenced by *Cuscuta*, suggests a hormonal influence (cf. ABOU-MANDOUR et al. 1968). MILTHORPE & MOORBY (1969) have stressed that it is not clear whether the stimulation by growth substances of the movement of metabolites is due to effects on movement within sieve tubes, entry or exit therefrom, or entry into associated parenchyma cells. *Cuscuta* could influence the rate of sink activity by a stimulating influence upon the exit of metabolites from the sieve tubes which could result in a stimulated long distance transport to the strong sink, possibly via a pressure-driven Münch-type mass flow, as discussed by, e.g., CRAFTS & CRISP (1971) and MACROBBIE (1971).

As PATE & GUNNING (1972) in an epilogue state that the issue still has to be settled as to whether the transfer cell does indeed function in a superlative manner in exchanging solutes with its extracellular environment, we can conclude that the transfer cells of *Cuscuta* indeed seem to function in a superlative manner in absorbing the huge quantity of assimilates released by the phloem to the free space in parasitised stem parts. *Cuscuta*, being attached only to a very localised part of the phloem system of the host which represents a minor part of the total phloem system, can withdraw almost all assimilates exported by the leaves of a host, interfering completely with the translocation to the fruits (WOLSWINKEL 1974).

Our data on washing out ^{14}C-solutes from the free space, combined with the anatomical data presented by Dörr, suggest a membrane-mediated transfer of solutes from sieve tubes via the free space to the absorptive hyphae of *Cuscuta*. Difficulties seem to be aroused in this context by data on transmission of viruses (e.g. BENNETT 1944 and 1956, HOSFORD 1967) and mycoplasmas (e.g. DIJKSTRA & LEE 1972) by *Cuscuta*. The membrane-mediated transfer should refer mainly to low molecular solutes, as was also stressed by DÖRR (1972). ESAU et al. (1967), KITAJIMA & LAURITIS (1969), and DAVISON (1969) observed passage of viruses through plasmodesmata. DÖRR (1972) mentioned as a possible route the plas-

modesmata of searching hyphae or the open plasma connections she very rarely observed after hyperinfection of the tissue of the host; in the last case she found the absorptive hyphae to penetrate to the sieve plate. ESAU et al. (1967) suggested the possibility that viruses may destroy the plasma membrane of a sieve tube and pass directly through the wall as complete or incomplete particles. In a review paper ESAU (1697) referred to studies in which it seems that viruses are able to pass through plasma membranes.

Apparently macromolecular particles may find another way than typical metabolites to penetrate into *Cuscuta*. The data presented support the idea that membrane-mediated transfer of solutes is essential for transfer to *Cuscuta*. The absorptive hyphae of *Cuscuta* can be placed in a much wider context in the light of the growing quantity of data on transfer cells. The data on the enhanced unloading of phloem may be meaningful for efforts to elucidate the normal movement of solutes in plants between phloem and surrounding tissues. Fruits, like *Cuscuta* characterised by a very intensive growth, form one example of a situation in which an enhanced unloading of phloem could be essential for the supply of assimilates to the strong sink.

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