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STUDIES ON THE ABSORPTION, DISTRIBUTION AND RELEASE OF LABELLED RUBIDIUM IONS IN YOUNG INTACT BARLEY PLANTS

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INTRODUCTION

Since the classic study on rubidium absorption by potato discs by STEWARD and HARRISON (1939), the rubidium ion has become more and more popular in plant physiology, especially in the field of active uptake studies. One reason for this is the absence of rubidium in plant material at the start of the experiments. Another one, nowadays, is the availability of a very useful radioactive isotope, which greatly facilitates determination and, in addition, renders it possible to study exchange phenomena. The most important aspect, however, is the similarity of the rubidium and potassium ions, so that much knowledge about the behaviour of the potassium ion can be gathered by using rubidium salts.

On the other hand, it is well-known from studies on plant nutrition that potassium cannot be substituted by rubidium. Quite recently SCOTT and DE VOE (1957) summarized the various findings on the diverging effects of potassium and rubidium in various physiological processes. In addition, they found that in *Ulva lactuca* replacement of potassium by rubidium differed from the reverse process. In experiments with intact barley plants by the present author it was found that uptake of labelled rubidium was much more inhibited by a pretreatment with rubidium than by a similar treatment with potassium. Clearly, care should be taken in comparing results obtained for rubidium with those obtained for potassium (MULLINS and BROOKS 1939, OVERSTREET and BROYER 1940, SUTCLIFFE 1954).

The experiments reported here, were started at the end of 1955. Much time was devoted to the study of the influence of various pretreatments on subsequent rubidium uptake (HELDER 1957). Here results have been selected on the time course of rubidium absorption, distribution and release. It should be emphasized that all experiments have been done with intact plants (SCOTT RUSSELL 1954).

MATERIAL AND METHODS

Young barley plants were grown on Hoagland solution and used while in the second leaf stage. In each experiment 8 sets of 12 plants were prepared and placed in the constant temperature room one day before the start of the experiment. The sets were placed on simple

earthware jars of half a liter capacity. In the majority of the experiments the plants were pretreated in water for about 18 hours, whereas absorption took place from .001 M labelled rubidium solution (10–20 μC/l). In both water and rubidium solution some calcium sulphate was present. The importance of this will be illustrated by some experiments in which the omission of calcium sulphate was studied.

The following standard procedure was applied to changing the solutions and harvesting the plant material (HELDER 1957). The old solution was removed and the roots allowed to drain for one minute. The roots were then washed for one minute in water containing some calcium sulphate in order to remove the adhering solution and were then allowed to drain for another minute. The plants were subsequently either harvested or placed in a fresh solution. By this procedure not only the adhering solution but also rubidium ions, loosely held by the roots were removed, as will be discussed later on.

The plant material was dried, weighed, ashed with a mixture of perchloric, sulphuric and nitric acid and the labelled rubidium content of roots and shoots separately determined.

EXPERIMENTS

The course of Rb content*

In contrast to what was expected, the rate of Rb* uptake appeared to be about constant in most experiments for at least the first six hours (Fig. 1). Of course, it took some time before the first amounts of Rb* were found in the shoots. As a result the increase in Rb* content of the shoots became higher after a certain lapse of time, whereas the reverse applied to the content of the roots. In experiments with shorter absorption intervals it was found that the transfer to the shoots may become constant after 1–2 hours.

Although there was no gradual slowing down of the uptake fig. 1

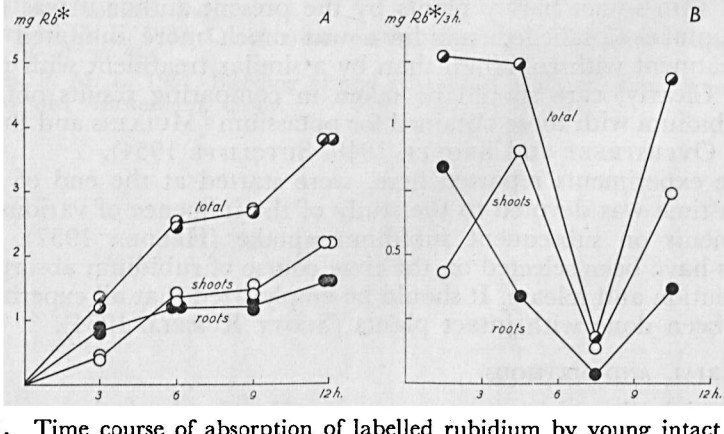

Fig. 1. Time course of absorption of labelled rubidium by young intact barley plants. Sets of 12 plants were allowed to absorb from a .001 M Rb*Cl + $CaSO_4$ solution for various periods. A: Total amounts present at the end of the absorption periods. B: Rate of increase of Rb* content in successive periods.

illustrates that there can be a more or less sudden depression in the absorption process, followed by a resumption of the uptake at about its initial speed. In this example there was a complete stand-still of the fixation of Rb* within the roots, whereas there remained a slight transfer to the shoots.

In other experiments a similar depression was observed, but not until after about 6 hours after the start of the experiment. Fig. 2 shows the results of an experiment in which maximum depression occurred from 9 to 12 hours after the start of the experiment. When comparing the values for rate of fixation within the roots and transfer to the shoots it should always be realized that there is a tendency for the shoot values to be low during the first absorption period for reasons already explained.

Clearly, carrying out these experiments necessitated that the various sets of plants had to absorb the Rb* at different times of the day. This means that the course of Rb* content found in the previous experiments might be related to periodicity phenomena as were observed by HANSON and BIDDULPH (1953). From the design of the various experiments on the course of Rb* content it could be deduced that

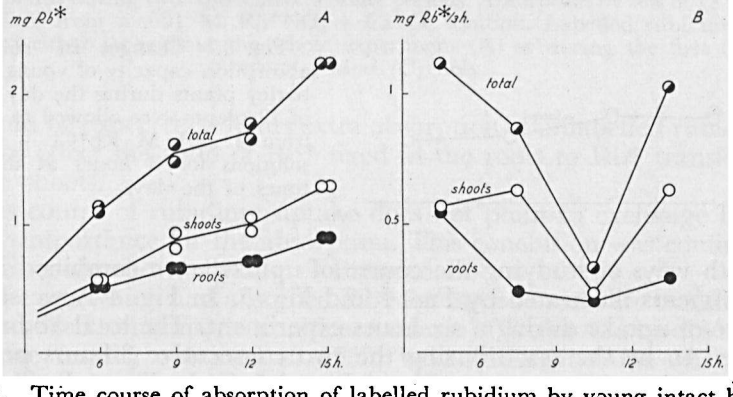

Fig. 2. Time course of absorption of labelled rubidium by young intact barley plants. Further details in fig. 1.

Rb absorption should have been low mainly from 3 p.m. till 6 p.m. if periodic fluctuations in absorption capacity were responsible for the depression. In Fig. 3 an experiment is given in which the amount of Rb* absorbed at various intervals during the day was estimated. There is a gradual decrease of the absorption capacity, although the moment at which this decrease started varied somewhat in different experiments. The decrease in the amounts transferred to the shoots was relatively larger than the decrease in the amounts found in the roots so that the transport-index dropped from about 27 % to 20 %.

Nevertheless, the lowest absorption values in these experiments were about 70 % of the highest value. Therefore, there can be little doubt that periodicity cannot account for the enormous depressions found in the previous experiments.

The time course can also be studied by putting sets of plants on

an unlabelled rubidium salt solution for various periods of time and then on a labelled solution for a definite period. In one particular experiment the plants were placed on a RbCl .001 M solution for either 0, 2, 4 or 6 hours and then allowed to absorb Rb* from a labelled solution of similar composition for 2 hours. The advantage of this design is that the amount found in roots and shoots can be compared directly as there had been no Rb* in the plants at the start of the final absorption period. Also in this experiment there was a constant rate of uptake after 0, 2 and 4 hours Rb absorption. After 6 hours absorption, however, a clear decrease occurred.

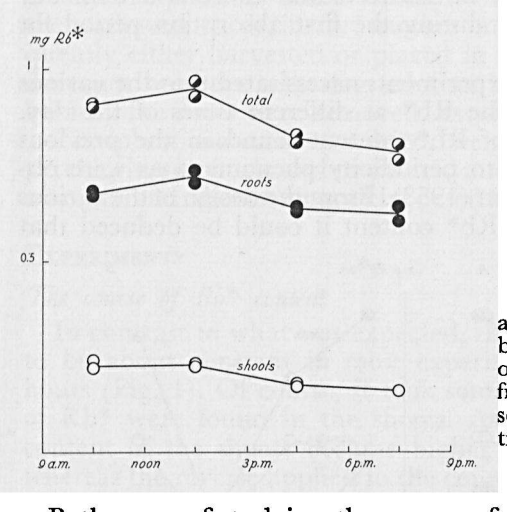

Fig. 3. Changes in rubidium absorption capacity of young intact barley plants during the day. Sets of 12 plants were allowed to absorb from a .001 M Rb*Cl + $CaSO_4$ solutions for 3 hours at different times of the day.

Both ways of studying the course of uptake were combined in the experiments illustrated by Fig. 4 and Fig. 5. In Fig. 4 A we see the course of uptake during a six hours experiment. The total absorption proved to be the same during the two consecutive 3-hours periods. This can also be seen from Fig. 4 B and 4 C. In Fig. 4 B the same absorption values at the end of the first three hours, already indicated in A, have been plotted at a somewhat larger scale. In C the values are given for sets of plants which had been on unlabelled solution during the first period and on a labelled solution during the second period only.

Obviously, there was no difference as to total amounts absorbed during the first and second period. The sum of both amounts was equal to the total amount absorbed by sets which had been on the labelled solution for six hours. The more striking is the change in distribution of the absorbed Rb* among roots and shoots. As appears from Fig. 4 B and 4 C the amounts taken up from the medium and transferred to the shoots were much higher during the second period (C) than during the first one (B). This shift in distribution took place without having any influence on the entrance of the Rb* into the plants. It points to the fact that ions are taken up by one process which is more or less independent from other processes which are involved

in the subsequent distribution of the absorbed rubidium ions among roots and shoots.

Fig. 5 shows the results of a similar experiment for plants which had been pretreated in unlabelled Rb solution for 24 hours. The results are essentially the same, except for the shift in distribution.

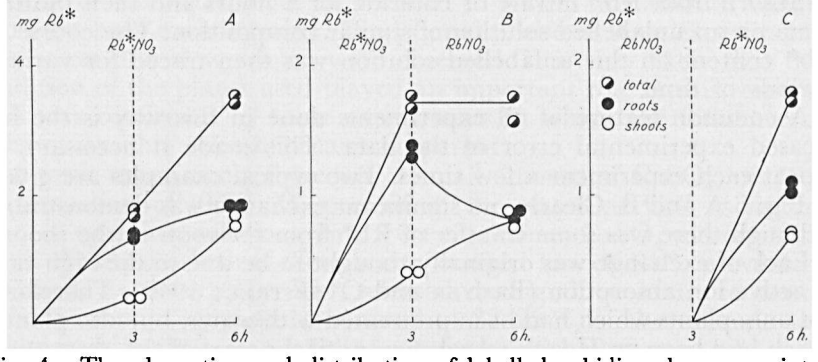

Fig. 4. The absorption and distribution of labelled rubidium by young intact barley plants during two consecutive 3 hours periods. Absorption by sets of 12 plants took place from a .001 M $Rb^*NO_3 + CaSO_4$ solution. Labelled rubidium was supplied either throughout the whole experiment (A) or during the first (B) or second period (C) only.

As could be expected, 3 hours extra absorption of unlabelled rubidium did not effect the ratio of Rb* fixed in the roots to Rb* transferred to the shoots.

This course of rubidium uptake does not point to exchange being of any importance for the absorption. This conclusion was confirmed by the results shown by Fig. 4 B and 5 B.

If the sets which had absorbed Rb* for three hours were transferred to unlabelled solutions, no significant release of Rb* could be observed. As these results, obtained in 1955, were different from what was normally assumed to be the case for cation absorption, many more

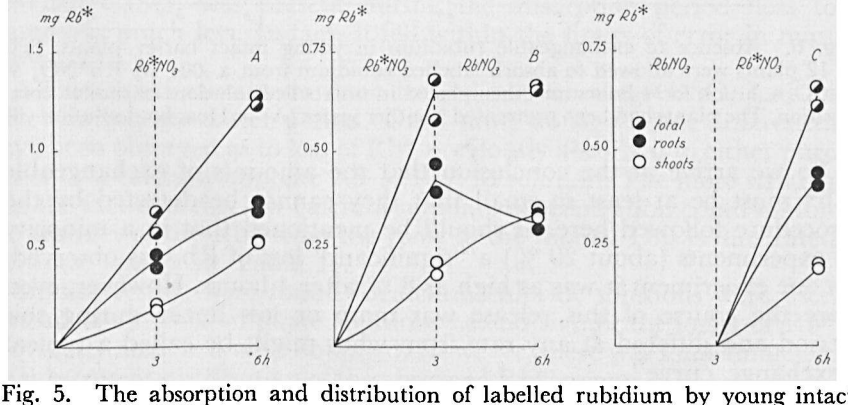

Fig. 5. The absorption and distribution of labelled rubidium by young intact barley plants during two consecutive 3 hours periods. The plants were pretreated in unlabelled rubidium solution for one day. Further details in fig. 4.

experiments were devoted to the study of the exchangeability of the absorbed Rb* ions.

Experiments on exchange of Rb previously absorbed*

Most of these experiments were done by allowing sets of plants to absorb from Rb* nitrate or chloride for 4 hours and then putting them on an unlabelled solution of similar composition. The course of Rb* content on this unlabelled solution was then traced for various periods of time.

A common feature of all experiments done in this way is the increased experimental error of the data. This made it necessary to repeat each experiment a few times. Two typical examples are given in Fig. 6 A and B. Clearly, no significant exchange was demonstrable although there was some transfer of Rb* from the roots to the shoots.

Lack of exchange was originally thought to be due to the high rate of active ion absorption (BROYER and OVERSTREET 1940). Therefore, not only plants which had been pretreated with water, but also plants, which had been on Hoagland solution until the start of the absorption period, were tested. The ability for absorbing Rb is influenced by these treatments to a large extent, in that after a water treatment absorption capacity is largely enhanced (HELDER, 1957). No differences were found, however, in regard to these exchange phenomena.

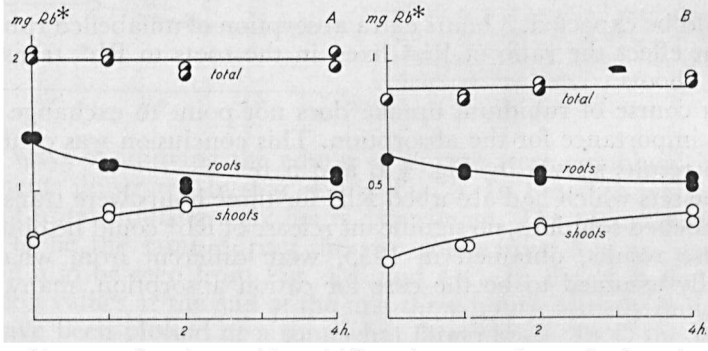

Fig. 6. Absence of exchangeable rubidium in young intact barley plants. Sets of 12 plants were allowed to absorb labelled rubidium from a .001 M Rb^*NO_3 + $CaSO_4$ solution for 4 hours and then placed in unlabelled solutions of similar composition. The plants had been pretreated in either water (A) or Hoagland solution (B)

So we arrive at the conclusion that the amounts of exchangeable Rb* must be at least so small that they cannot be detected by the procedure followed here. It should be mentioned that in a minority of experiments (about 20 %) a "significant" loss of Rb was observed. In one experiment it was as high as 8 % after 4 hours. However, even here the course of this release was more or less linear during this period and differed at any rate from what might be called a typical "exchange curve".

Loss of Rb, previously absorbed, to various solutions*

Instead of tracing the release of Rb*, absorbed from labelled

solutions to unlabelled solutions of the same composition, this can also be done using solutions which differ from the labelled solutions.

If sets of plants were transferred to water there proved to be little or no loss of the Rb* previously absorbed. This could be expected on the basis of the results on exchange, for, if there was an appreciable amount of Rb* that could be released to water, this amount would also have been exchangeable. Exceptionally, also in these experiments some cases of a slow release were observed. It is very likely that the condition of the plants used played an important role in this respect. This was confirmed by some experiments in which the influence of calcium on this release became apparent.

In most experiments there was a small amount of $CaSO_4$, up to a concentration of .0005 M, present in the water as well as in the labelled rubidium solutions. In a few experiments $CaSO_4$ was omitted. In all these experiments there was a marked loss of Rb*, previously absorbed, to water. The Rb* released during three hours to water without calcium sulphate amounted to 22–25 % of the amount absorbed previously from a labelled solution for 2 hours, as can be seen from Table I.

TABLE I

The loss of labelled rubidium, previously absorbed, to water.

Sets of 12 young barley plants were allowed to absorb rubidium from a .005 M Rb* Cl solution without $CaSO_4$ for 2 hours and placed subsequently in demineralized water for 3 hours.

Rb* content		Loss of Rb* in percentages of initial content
at the end of absorption-period	at the end of water treatment	
.82 mg Rb*	.64 mg Rb*	23 %
1.22 mg Rb*	.93 mg Rb*	24 %
1.59 mg Rb*	1.19 mg Rb*	25 %
1.20 mg Rb*	.94 mg Rb*	22 %

When $CaSO_4$ was present during the absorption period, loss to water was much less. In fact, it fell within the limits of error in most cases.

A few experiments were done to find out whether $CaSO_4$ had an effect during the water period. Until now, no significant differences have been observed as to loss of Rb* previously absorbed to either pure water or a calcium sulphate 2.5×10^{-4} M solution. The more striking was the fact that this low calcium sulphate concentration could inhibit the transfer of the Rb* from the roots to the shoots. This is illustrated by some figures in Table II.

Similar results were found if calcium chloride solutions were used instead of calcium sulphate solutions i.e. no significant losses of Rb* previously absorbed were obtained. However, there was some influence on subsequent distribution of the absorbed Rb* among roots and shoots in that slightly more Rb* was transferred to the shoots if the plants were on calcium chloride. The highest rate of transfer was, however,

always obtained in unlabelled rubidium solutions. Clearly, both the anion and the cation have some bearing on the process of release of Rb* to the xylem vessels.

Quite another picture was obtained with rubidium sulphate. If plants were placed on a labelled rubidium nitrate solution for 4 hours

TABLE II

The transfer of labelled rubidium, previously absorbed, from roots to shoots, as influenced by various solutions.

Sets of 12 intact barley plants were allowed to absorb from .001 M Rb* Cl + $CaSO_4$ for two hours and then placed in various unlabelled solutions for 3 hours.

Rb* content of the shoots at the end of the absorption period	Subsequent increase of Rb* content of the shoots	
	water	$CaSO_4$
.23 mg Rb*	+ .09 mg Rb*	+ .07 mg Rb*
.22 mg Rb*	+ .17 mg Rb*	+ .14 mg Rb*
	$CaSO_4$	$CaCl_2$
.16 mg Rb*	+ .08 mg Rb*	+ .16 mg Rb*
.14 mg Rb*	+ .11 mg Rb*	+ .14 mg Rb*
	$CaCl_2$	RbCl
.12 mg Rb*	+ .14 mg Rb*	+ .16 mg Rb*
.13 mg Rb*	+ .11 mg Rb*	+ .14 mg Rb*

and then transferred to unlabelled rubidium sulphate solution, significant amounts of Rb* (mostly of the order of 20–25 %) were released. Also in these experiments the variability of the figures may be rather high, but the release observed is beyond any doubt. One of the best results obtained is shown by Fig. 7, demonstrating the course of Rb* content on a rubidium sulphate solution. It should be noticed that during the period of rapid release there was very little transfer, if any, to the shoots.

At first sight these experiments seemed to show an exchange of Rb* for Rb. The presence of exchange here and the lack of exchange in the experiments discussed in the previous section might be explained by assuming a strong reduction of active uptake in the case of the rubi-

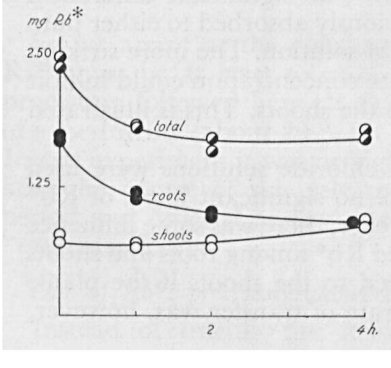

Fig. 7. Loss of labelled rubidium, previously absorbed by young intact barley plants to unlabelled rubidium sulphate solution ("apparent exchange"). Sets of 12 plants were allowed to absorb from a .001 M Rb^*NO_3 + $CaSO_4$ solution for 4 hours and then placed in a .0005 M $RbSO_4$ + $CaSO_4$ solution for various periods of time.

dium sulphate solution. In fact, it was found that the rate of Rb absorption from a sulphate solution was only one fifth of that from a nitrate solution.

However, a check was made by putting sets of plants on an unlabelled rubidium nitrate solution first and then on a labelled rubidium sulphate solution. The labelled rubidium absorbed from the sulphate solution in four hours amounted to .24 mg, whereas the labelled rubidium absorbed from the nitrate solution had been 3.88 mg and dropped to 2.92 mg after a stay on the unlabelled sulphate solution. Clearly, the amounts given off (.96 mg) exceeded the amounts taken up (.24 mg), so that exchange could only partly account for the losses observed.

Similar effects were observed in experiments with potassium sulphate. However, other potassium salts also proved to be able to remove part of the Rb* absorbed. The results of two experiments are given in Table III.

TABLE III

Changes in labelled rubidium content of roots and shoots of young barley plants as influenced by various unlabelled solutions.

Sets of 12 plants were allowed to absorb rubidium from a .001 M Rb* NO_3 solution with $CaSO_4$ for 4 hours and placed subsequently in .001 M unlabelled solutions of either rubidium or potassium or calcium nitrate.

Rb* content at the end of absorption period		Changes in Rb* content during subsequent treatment on		
		$RbNO_3$	KNO_3	$Ca(NO_3)_2$
Experiment A				
roots	1.42 mg Rb*	— .42 mg Rb*	— .42 mg Rb*	— .39 mg Rb*
shoots	.59 mg Rb*	+ .35 mg Rb*	+ .19 mg Rb*	+ .33 mg Rb*
	2.01 mg Rb*	— .07 mg Rb*	— .23 mg Rb*	— .06 mg Rb*
Experiment B				
roots	1.30 mg Rb*	— .36 mg Rb*	— .36 mg Rb*	— .29 mg Rb*
shoots	.55 mg Rb*	+ .36 mg Rb*	+ .18 mg Rb*	+ .29 mg Rb*
	1.85 mg Rb*	.00 mg Rb*	.17 mg Rb*	.00 mg Rb*

The small losses by the whole plants observed in experiment A, for Rb and Ca nitrate were not significant. Clearly, large amounts were lost to the potassium solutions. On the other hand, the amounts transferred to the shoots are much less in this case, but the final content of the roots was about the same for Rb and K nitrate; the main difference being that in Rb nitrate the labelled rubidium is moved upwards to the shoots, whereas in K nitrate about one half is lost to the medium instead of being transferred to the shoots. It would have been interesting to know how much K was transferred to the shoots concomittantly.

If transfer to the shoots of the sets in the Rb nitrate solution is compared with that of the sets in the calcium nitrate solution a slight depression of this transfer in sets standing on calcium nitrate could be observed. This results corroborates the former ones obtained with rubidium and calcium chloride.

Discussion

In the experiments on the course of rubidium absorption we observed more or less sudden decreases in the rate of uptake. As external conditions were kept constant, these decreases must be ascribed to some internal factor. It was found that variations in absorption, due to autonomic periodicity, were too slight to account for the enormous depression often obtained. As the phenomena were obtained only after the plants having been on a rubidium solution for at least 6 hours, it seems reasonable to suggest that the rubidium ion is itself responsible for these effects. During the last few years striking effects of various ion species on salt absorption and protoplasmic streaming have been observed in our laboratory (Helder 1957, Jager 1958, Sol 1958).

The rubidium absorbed is partly transferred to the shoots. In short periods the amounts of rubidium present in the shoots are bound to be small because of the fact that it takes some time before the first amounts have passed through the root tissue and xylem vessels upwards to the shoots. However, it was also found that during the first few hours more rubidium was fixed than afterwards. This became apparent from experiments in which plants were placed on unlabelled rubidium solutions prior to the absorption period proper. Various explanations are possible. The most likely one is, that the rubidium ions which had entered the roots were used up by separate processes which fixed the ions in the root tissue. If one of these processes diminished because of saturation, more ions would become available for upward movement to the shoots.

It is remarkable that this shift in distribution could occur without having any influence on the rate of absorption from the medium. It points to the fact that this entrance of the ions into the root tissue is independent of what is going to happen with the ions afterwards, and that distinct processes were involved in the absorption, fixation and transfer of these ions.

According to current ideas the rate of the rubidium uptake could be expected to be fast at the beginning and to slow down regularly until a steady level was reached. Extrapolating the curve back to zero time would then have rendered possible a calculation of an initial rapid, most likely physical, uptake. This initial uptake could have been expressed in terms of apparent free space (Briggs 1957). In fact, in most experiments uptake was constant from the very beginning for at least the first 6 hours. At first sight these results were somewhat surprising. However, various aspects should be taken into account.

If we assume an apparent free space of the order of 25 % the amount of rubidium present in the free space could be calculated to be of the order of 20–25 μg if the plants were placed on a .001 M rubidium solution. Clearly, these amounts could hardly be detected under the conditions of very rapid active accumulation of these experiments. In experiments, which have emerged from these ones (van der Heide, Brouwer), it was proved that at higher external rubidium concentra-

tions more significant values could be obtained. But, what is more, the type of the plant system used for these studies proved to be very important, in that higher values for the apparent free space were found for decapitated plants than for intact plants, but the highest values were for submerged root systems.

Finally, it should be realized that in all experiments presented in this paper, the roots were rinsed for one minute before harvesting. In order to determine how much rubidium was removed by this procedure plants were allowed to equilibrate in a 5×10^{-4} M rubidium solution and then allowed to absorb from a fresh solution of similar composition. Absorption was estimated by determination of the drop in concentration. In addition, the solution was drained off, the roots rinsed for one minute according to the standard procedure and the absorption again estimated by determining the whole amount of rubidium left after being collected in this way. The uptake, as determined according to the last method, was lower. This proved that by rinsing the roots not only solution adhering to the roots had been removed, but also some rubidium attached to the roots. The amounts in terms of apparent free space, could be as high as 300–400 %. Most likely, these amounts represent rubidium ions which were adsorbed very loosely to the root surfaces.

What has been said about the apparent free space in relation to the time course of rubidium absorption also applies to the results of the experiments in which plants were changed from a labelled rubidium solution to water. In the majority of these experiments no significant losses were obtained. However, it was found that large quantities could be observed to be lost, in those cases in which previous uptake had taken place in the absence of calcium (compare OVERSTREET, JACOBSON and HANDLEY 1952). These results are reminiscent of the classical studies on the beneficial effects of balanced salt solutions in particular of calcium containing solutions, on the structural properties of the protoplasm (FISCHER 1956, HELDER 1956). Overstreet et al. ascribed their results to a higher stability of the rubidium binding entities in the cytoplasm.

It should also be realized that there may be differences between intact plants and immersed excised root systems. The presence of calcium during the water treatment did not seem to have any obvious influence on the loss of rubidium to the medium, but it could have a slight influence on the upward movement of the rubidium previously absorbed. This suggests that in the absence of calcium, more free rubidium ions are present in the roots and available for release. In addition there must be some effect determining whether these liberated rubidium ions are given off to the medium or not.

This view is nicely supported by the results of the experiments with unlabelled solutions of rubidium sulphate and potassium sulphate and nitrate. On the sulphate solutions there was a loss of previously absorbed rubidium to the medium. While this loss was going on, hardly any rubidium was translocated to the shoots. In potassium nitrate about the same quantities were left in the root tissue as in

unlabelled rubidium nitrate solution, the only difference being that in rubidium nitrate the labelled rubidium had been transferred to the shoots instead of to the medium.

Why it is that some solutions could bring about this release to the medium is difficult to say. This is particularly so when it is realized that rubidium sulphate and potassium nitrate did induce this release, but rubidium nitrate did not. Such a release, however, seems often to be associated with any change of solution. Even if an old solution is changed for a fresh one of similar composition a release can be induced which gradually changes into a reabsorption (ARISZ 1958). These phenomena may be ascribed to readjustments of the cytoplasm to new external conditions, but, clearly, this does not add very much to our understanding of the changes involved. On the other hand, it can be doubted whether the significance of these phenomena is sufficiently appreciated in studies on the apparent free space.

The results obtained with rubidium sulphate were also very instructive from another point of view. At first sight they seemed to indicate a ready exchange of labelled ions of the roots for unlabelled ions from the medium. However, there proved to be hardly any exchange, as the amounts released proved to be much greater than the amounts taken up simultaneously. In the opinion of the author too often exchange has been studied merely by comparing the release of labelled ions, previously absorbed, to unlabelled salt solutions and to water.

The results obtained here, also point to a difficulty in exchange studies. For instance if it is observed that the release and the simultaneous uptake of another ion are about equal, one is inclined to ascribe the ion fluxes to "exchange". Admittedly, no objection can be raised if the term "exchange" is only used to describe what has been observed. But it should be realized that the processes underlying this exchange may be quite different from simple exchange phenomena known from physico-chemical systems (RUSSELL and AYLAND 1955).

Anyhow, the exchangeable fraction of the rubidium ion in the root tissue is quite unimportant if compared with the fraction that can be induced to be transferred subsequently to the shoots. Of course, part of this "mobile" rubidium will also become more definitely fixed within the roots themselves. Therefore, a competition between both processes, fixation within the roots and transfer to the shoots can be expected. A few factors were found to have an influence on this competition. The distribution of the rubidium, previously absorbed, was found to depend on the composition of the external solution. At this stage it is difficult to say in what way the various ions caused the effects observed. If there is a store of loosely held rubidium ions, which is in equilibrium with the free mobile ions, external factors may have an influence on the equilibrium, increasing or decreasing in this way the amounts available for final distribution. However the processes of accumulation in the vacuoles and the release of the ions to the xylem vessels may be influenced. As both processes compete for the same ions, it is sufficient for a factor to act on both processes in a

similar but quantitatively different way. For instance calcium sulphate, if compared with water retarded subsequent transfer of rubidium to the shoots. It is possible that it either inhibited the release of ions to the xylem vessels or stimulated fixation within the roots or stimulated both processes but the fixation within the roots to a higher degree. Also an influence on the equilibrium, mentioned above is conceivable.

Most interesting was the stimulating effect of calcium chloride on the subsequent transfer. This must be due to the chloride ions as calcium can be expected to hamper rather than stimulate the transfer. It is known from exudation studies that as soon as chloride is withdrawn from the external solution, the secretion of ions to the xylem vessel is decreased almost instantaneously (salt effect-VAN ANDEL 1953). The reverse applies for addition of chloride ions. Therefore, the most likely explanation for the higher rate of transfer found in these experiments seems to be a facilitated release of rubidium ions to the xylem vessel caused by the continued release of chloride ions. Such a mutual effect of cations and anions is well-known for active absorption and it seems not unreasonable to apply it to the process of ion release.

An increased transfer to the shoots if the plants are transferred to a rubidium chloride instead of a calcium chloride solution is not very surprising. But also here, various explanations are possible. On calcium chloride the mobile rubidium fraction will disappear gradually. This may increase the ratio of ions fixed in the roots to ions transferred to the shoots as the root cells have first access to these ions. However, it is also possible that, owing to an exchange of newly introduced unlabelled rubidium ions for labelled ions loosely held by the cytoplasm, the number of labelled ions available for fixation and transfer is increased.

It cannot be denied that following these lines of thought the whole picture of uptake, fixation and transfer of rubidium ions by intact barley plants becomes rather complicated. Indeed, it is quite different from the simpler picture according to which ions are introduced into a free space and passively carried by the transpiration stream through the root tissue into the xylem vessels, the only active process being the accumulation in the vacuoles (EPSTEIN 1957). The main reason for this may be that the first picture was arrived at starting from experiments with whole plants, while many ideas on the absorption process are based on results with tissue slices and excised root systems only. It has already been emphasized that the use of different plant systems may lead to divergent results and conclusions. On the other hand, it is readily admitted that much work remains to be done on the various aspects of absorption and distribution of ions in intact plants, touched upon in this discussion.

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SUMMARY

Young intact barley plants proved to absorb labelled rubidium at an almost constant speed during the first six hours of an experiment. Afterwards severe depressions in the absorption ability occurred followed by a resumption of the uptake at about its initial speed. Only a very small fraction, if any, of the rubidium absorbed could have been exchangeable, as no significant loss of labelled rubidium to unlabelled solutions was detected. However, on rubidium sulphate, potassium sulphate and potassium nitrate solutions marked amounts of labelled rubidium, previously absorbed, were given off. A loss to water could also be observed, provided previous rubidium absorption had taken place in the absence of calcium sulphate. The ionic composition of the medium also had an influence on the transfer of rubidium ions to the shoots.

It was assumed that at any moment part of the rubidium ions in the symplasm of the roots are in a mobile state, i.e. they can be either more definitely fixed within the root tissue, or transferred to the shoots or exceptionally given back to the medium. The external solution was thought to have an influence on the total amount of mobile rubidium ions, on the structural features of the symplasm (permeability) and on its functional features (active absorption into the symplasm, fixation in the symplasm, secretion to the vacuoles, and transfer to the shoots).

Stress was laid on the importance of the ionic composition of the medium and the plant system used for studies on free space and exchange phenomena. It was found necessary to postulate a complex physiological system relating the absorption and distribution of rubidium in intact plants, in order to be able to account for the divergent effects observed.

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