THE ABSORPTION OF THALLIUM IONS BY EXCISED BARLEY ROOTS

G. G. J. BANGE AND F. VAN IREN

Botanisch Laboratorium, Leiden

SUMMARY

The kinetics of Tl absorption by excised barley roots were studied with respect to the time and concentration dependence, sensitivity to Ca, and interaction with Rb and Na.

The following conclusions were drawn:

1. Tl is bound by sites normally binding K.
2. In the range of concentrations from 0 to 0.2 me/l, Tl is absorbed readily into the protoplasm but vacuolar accumulation is slow and tends to stagnate.
3. Although O_2 uptake is progressively reduced in the presence of Tl, this effect does not seem responsible for the stagnation of vacuolar absorption.
4. Besides a competitive inhibition, there is a stimulation of vacuolar Tl transport by Rb but not by Na.

The peculiarities of Tl absorption are tentatively explained on the basis of structural interactions between a macromolecular carrier and the ions transported.

1. INTRODUCTION

Study of the absorption characteristics of such elements as Rb, Cs, Sr, or radicals such as SeO_4, Cr_2O_4, AsO_4 – which to varying degrees might be called 'physiological isotopes' of the major nutrient elements (K, Ca) or radicals (SO_4, PO_4) – has been a useful tool in elucidating several aspects of the process of ion transport in the living cell. The term 'physiological isotopes', however, is misleading in that it implies severe underestimation of the discriminative faculty of the living system toward these ions or ionic groups. Actually – and otherwise than in the work with chemical isotopes – it is these discriminating features that can be made use of in studies on ion transport.

This paper reports the results of some experiments on the ability of plant roots to absorb and accumulate monovalent Tl ions. COHEN (1962) has presented evidence that Tl ions have an extremely high affinity for K-specific binding sites in *Chlorella*. Furthermore, Tl appears to be a very efficient substitute for K in activating the (Na + K) dependent ATP-ase of animal tissues (BRITTEN & BLANK 1968).

With these facts in mind it seemed interesting to establish the extent to which Tl ions can serve as a substitute for K ions in the absorption process in barley roots and in which respects they fail to do so.

2. MATERIAL AND METHODS

Seeds of *Hordeum vulgare* L. cultivar "Herta" were disinfected in a 1% $HgCl_2$ solution for one minute, rinsed in running tap water for about 20 minutes, and allowed to germinate in fine sand moistened with tap water for about 24 hours. After this period they were separated from the sand by sieving. The seedlings were raised on a dilute $CaSO_4$ solution (2×10^{-4} M) in the dark at 25 °C as described by HOOYMANS (1964), whose procedures were also followed in the excision of the roots and their preparation for the experiments.

All absorption experiments were performed in polyethylene bottles under aeration with compressed air and at a temperature of 25 °C. Further conditions for the separate experiments are given with the figures.

The influence of Tl on cell respiration was studied by means of the Warburg technique. We had to compromise between the conflicting demands of, on the one hand, a sufficiently accurate estimation of O_2 consumption which benefits from a larger amount of roots, and on the other hand the maintenance of the rate of Tl absorption, which does not permit a strong depletion of the Tl solution and thus requires a smaller amount of roots.

Tl absorption was estimated by analysis of either the root material or the experimental solution, as indicated in the separate experiments. Root analysis was performed by extraction of the roots with hot nitric acid (about 0.5 M) for about one hour. After paper filtration, the extract was made up to volume and its Tl content estimated in a Beckman model DU flame spectrophotometer at a wavelength of 377.5 mμ, by comparison with a series of standard solutions. Interference by other components of the extract was found to be negligible. The experimental solutions were analysed by direct flame-photometry with the exception of experiment 1 B, in which the following colorimetric procedure was used. To 10 ml aliquots of Tl_2SO_4 solution in the concentration range from 0.02 to 0.10 me/l Tl, 10 ml $NaHCO_3$ solution (6 mM) and 1 ml of a saturated Na_2S solution were added. Exactly 3 minutes after the addition of Na_2S, the intensity of the colour was compared to a series of standard solutions at a wavelength of 400 mμ.

Ion concentrations are expressed in milliequivalents per litre (me/l).

3. RESULTS

As a starting point for further kinetic experiments, the relation between Tl absorption and time was determined. As shown in *fig. 1A* (see also *figs 1B, 5A*, and *7A*) Tl uptake from a concentration of 0.2 me/l starts at a rate only slightly lower than that of the rapid initial phase discernible in the uptake of other monovalent cations such as Rb or Na (see *figs. 6A, 3A*, and *3B*) but after 3 hours the rate of Tl uptake becomes low and tends to decline further, which contrasts sharply with the linear and relatively steep uptake lines of Rb and Na in the second phase. Thus, after 3 hours the rate of Tl uptake is about 0.75 me/kg fresh weight per hour and after 6 hours it is as low as 0.25 me/kg fresh weight per hour. The strong inhibition of the uptake at low temperature (*fig. 1B*) in-

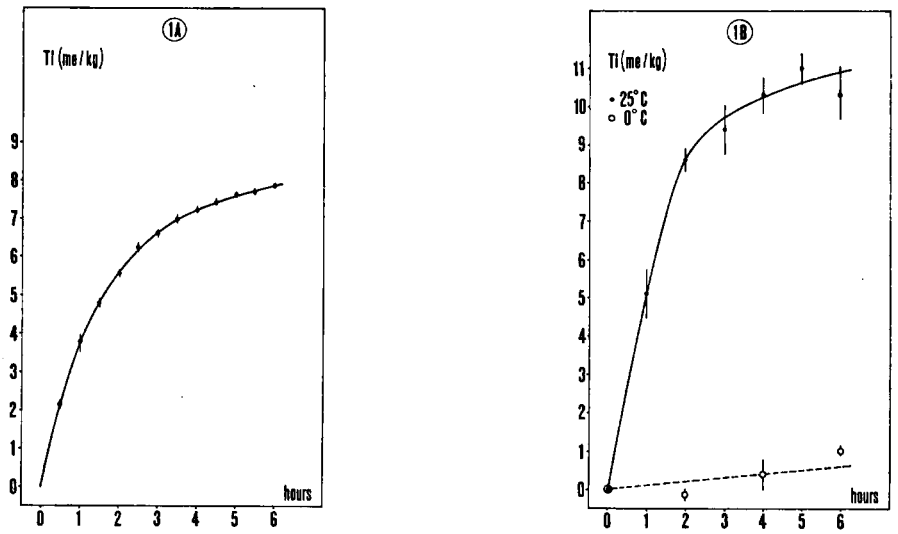


Fig. 1. A. Relation between Tl absorption and time at 25 °C. Eight grams of roots were allowed to absorb Tl from a 0.1 mM Tl_2SO_4 solution to which 5 mM $CaSO_4$ and 0.1 mM $Ca(HCO_3)_2$ had been added (pH ± 7). The volumes used varied from 400 ml at the start to 100 ml toward the end of the experiment. At half-hour intervals the uptake solution was decanted and renewed after a preliminary rinse with the same solution. The decrease in Tl content of the uptake solutions was determined by flame-photometry. The experiment was run in triplicate.
B. Comparison of the relation between Tl absorption and time at 25 °C and at 0 °C. Batches of 2.5 grams of roots were allowed to absorb Tl from 2 litres of a 0.05 mM Tl_2SO_4 solution to which 1 mM $CaBr_2$ and 0.1 mM $Ca(HCO_3)_2$ had been added (pH ± 7). At one (25 °C) or two (0 °C) hour intervals the uptake solution was decanted and renewed after a preliminary rinse with the same solution. The decrease in Tl content of the uptake solutions was determined colorimetrically. The experiment was run in duplicate.

dicates that the process is under metabolic control.

Because Tl is known to be poisonous to living organisms, it was essential to establish whether the low and still decreasing absorption rate in the second half of the experiment could be due to the absorbed ion interfering with general cell metabolism. The rate of O_2 consumption was used as an indication of the presence of such an effect.

Fig. 2 shows that the presence of a Tl concentration of 0.8 me/l induces a considerable deviation from the O_2 uptake of the control, which except for the first 1½ hours is linear. After 6 hours the rate of O_2 consumption has been lowered to about 35% of its original value (reference line). Roughly the same inhibition was found at a Tl concentration of 0.2 me/l. So Tl distinctly interferes with cell respiration; but further investigation is required to elucidate whether this is the direct cause of the low rate of Tl absorption after about 3 hours.

The best method to determine whether there is such a causal relation seems

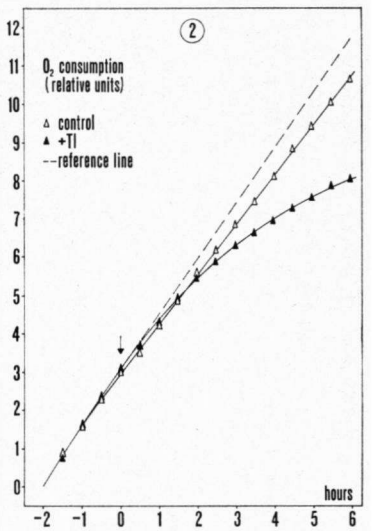


Fig. 2. The effect of Tl on O_2 consumption. Ten apical root segments of 5 cm each (± 125 mg) were cut in two and suspended in a Warburg-vessel in 1.5 ml of a basic solution of 4.5 mM $CaSO_4$ and 0.5 mM $CaCl_2$. After two hours (arrow), in 2 of the 4 vessels, 0.5 ml of basic solution to which 2 mM Tl_2SO_4 had been added, was poured onto the root suspension from the side-vessel; in the remaining 2 vessels only basic solution was added (controls). The initial Tl concentration of 1 me/l was estimated to be about halved after 3 hours of uptake. The points in the graph represent the mean of the replicates.

to consist in establishing the effect of the Tl-induced reduction in O_2 consumption on the simultaneous uptake of another monovalent cation. Several experiments on the effect of Tl on Na absorption in the absence of Ca were run. Na absorption proved to be considerably depressed by Tl, as it is by K (BANGE *et al.* 1965), though the result was variable with respect both to the degree of the inhibition of overall Na uptake and to the elimination of the rapid initial phase, as illustrated, for instance, by *figs. 3, A* and *B*, and *7C*. But from *fig. 3A* it is clear that Na absorption in the presence of Tl does not necessarily show a gradual decline in rate at all. On the other hand, in the experiment shown in *fig. 3B* there is a decline after 2 to 3 hours but the linearity as well as the relative magnitude of subsequent Na uptake may be regarded as proof that here the normal biphasic character of Na absorption is involved. A control experiment revealed that the presence of 0.2 me/l Na does not relieve the inhibition of O_2 consumption caused by Tl.

As far as the evidence goes, therefore, a disturbing effect of Tl on general cell metabolism, as indicated by the reduction in O_2 consumption, does not seem responsible for the slowing down of the uptake after the initial 3 hours, though the rather sharp drop in Na content after about 6 hours in *fig. 3A* may well have been due to Tl ultimately becoming deleterious to normal cell function.

The next experiment was designed to examine whether the absorption of Tl, like that of other monovalent cations, shows saturation kinetics, and how the uptake responds to the presence of excess Ca. It is called to mind here that according to HOOYMANS (1964) the response of the rapid initial phase of absorption to excess Ca (10 me/l) is dependent on the nature and concentration of the cation concerned: in the case of K (Rb), sensitivity to Ca was only apparent at the lowest K (Rb) concentrations (on the order of 10^{-2} me/l and lower), whereas

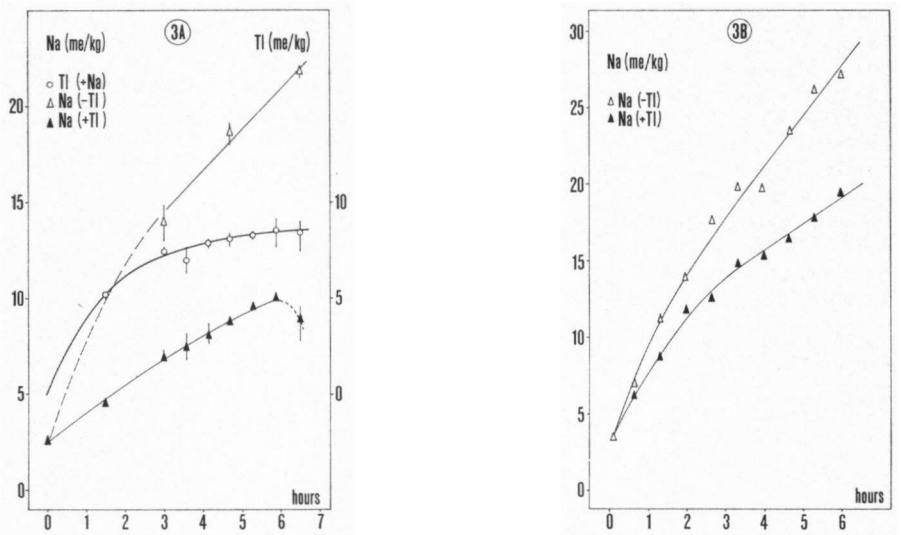


Fig. 3, A and B. Na absorption in the absence and presence of Tl. Batches of 3 grams of roots were suspended each in two or five litres of a 0.2 mM $NaHCO_3$ solution (pH ± 7.1) with or without the addition of 0.1 mM Tl_2SO_4. Root analysis was by flame-photometry. The first experiment (fig. 3A) was run in triplicate; the other (fig. 3 B) was a haplo-experiment.

for Na there was still a small Ca effect at 5 me/l Na. The relation between Tl absorption and concentration in the absence and presence of 5mM $CaSO_4$ is shown in *fig. 4*. Because of the decrease of the absorption rate with time (*cf. fig. 1*) it is necessary to be certain that the influence of concentration on the uptake rate is independent of the length of the absorption period. Comparison of the curves for 1 and 2 hours of absorption shows that this condition is satisfactorily fulfilled. Both curves for uptake in the absence of Ca have the form of ideal

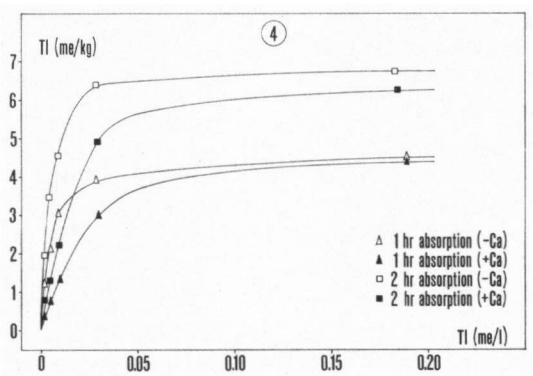


Fig. 4. Relation between the amount of Tl absorbed in one and two hours and the external Tl concentration. Batches of 1 gram of roots were immersed each for one or two hours in different concentrations of Tl_2SO_4 with and without the addition of 5 mM $CaSO_4$. The amount of experimental solution varied from 200 ml at the highest Tl concentration to 5 litres at the lowest. An estimated value for the pH, which was not under control in this experiment, is 6, but may have been somewhat lower (5.5) at the highest Tl concentration. Root analysis was by flame-photometry. No replicate of this experiment was run.

Langmuir adsorption isotherms as revealed by a Hofstee plot. The half-values estimated from this plot (method of least squares) are 0.0054 and 0.0047 me/l for 1 and 2 hours absorption, respectively. In the presence of Ca the half-value is raised to about 0.019 me/l, Tl uptake from the lowest concentrations being inhibited up to about 65% by Ca.

Thus, the response of Tl uptake to concentration and to the presence of excess Ca appears to fit in with the general pattern of monovalent cation uptake. The low half-value as well as the relative decrease of the Ca effect when Tl absorption reaches saturation (*cf.* HOOYMANS 1964) suggest that Tl, like Rb and Cs, makes use of sites normally binding K. In this case a mutual competitive inhibition must exist in the absorption of Tl and K (or Rb), though deviations from the ideal pattern may be anticipated from the large difference in their uptake behaviour after 3 hours.

Two experiments were performed to test this assumption. In the first (*fig. 5, A* and *C*), the influence of increasing amounts of Rb on Tl uptake from a 0.05 mM Tl_2SO_4 solution was studied. The remarkable outcome was that under these circumstances the total Tl content of the tissue after 7 hours first increases under the influence of small Rb concentrations before falling at higher Rb additions (*fig. 5A*). On the other hand, the amount of Tl absorbed after $1\frac{1}{2}$ hours decreased continuously. A close examination of the shape of the time curves reveals that this phenomenon is due to a different response to Rb of the rapid initial Tl absorption on the one hand and the slow or stagnant Tl uptake after 3 hours on the other. Whereas the former is increasingly inhibited by Rb, in the effect of Rb on the latter a stimulating as well as a competitive component can be recognized. In the presence of 0.25 me/l Rb, Tl is absorbed at a substantial rate after 3 hours. At higher Rb concentrations this rate is depressed again, but even in the presence of 1.25 me/l Rb, Tl uptake in the second phase is still more rapid than in the absence of Rb. At the same time, the decline in the rate of Tl uptake after 3 hours disappears gradually. *Fig. 5 C* shows that in concomitant Rb absorption the inhibiting effect of Tl in both uptake phases is overcome more and more at higher Rb concentrations, though a decrease in the rate of Rb uptake in the second phase seems to persist.

In the second experiment (*fig. 6, A* and *C*) the Rb concentration was kept constant at 0.2 me/l, whereas Tl concentration was varied from 0 to 0.5 me/l. In both phases of Rb absorption (*fig. 6 A*) the inhibition by Tl is apparent. At 0.3 me/l Tl, uptake of Rb has ceased after 3 hours and the initial phase still persisting is reduced further at 0.54 me/l Tl. Again at 0.18 me/l Tl, uptake of Rb after 3 hours seems to diminish gradually. Concomitant Tl uptake (fig. *6 C*) shows some paradoxical features. Whereas the rapid initial phase increases with rising Tl concentration, total uptake after $6\frac{1}{3}$ hours is lowest at the highest Tl concentration used. Analogous to the situation in *fig. 5A*, this phenomenon is clearly due to a different response of the rapid initial phase on the one hand and of the absorption after 3 hours, on the other, to a change in the ratio Tl/Rb. When this ratio is low, Tl uptake in the second phase is high and steady, but at higher ratios Tl is absorbed in this phase at a lower and declining rate. So, as in

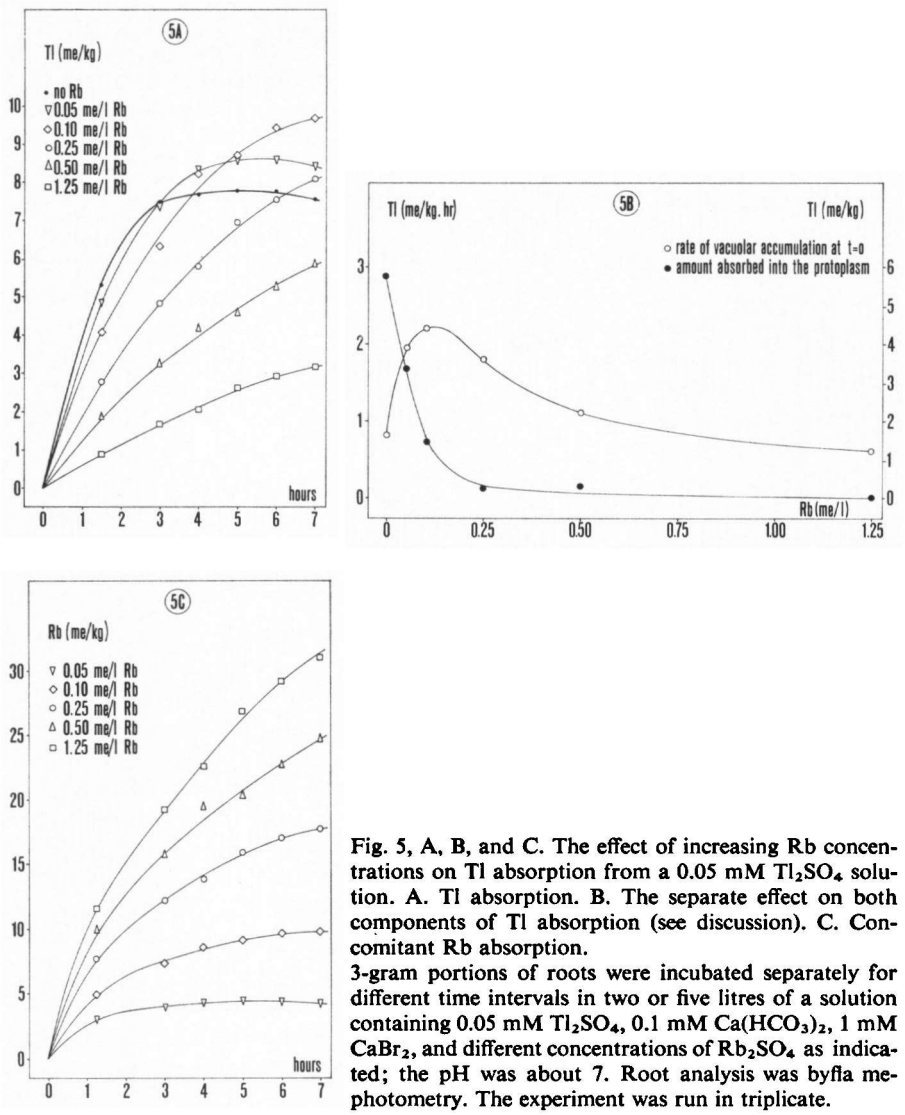


Fig. 5, A, B, and C. The effect of increasing Rb concentrations on Tl absorption from a 0.05 mM Tl_2SO_4 solution. A. Tl absorption. B. The separate effect on both components of Tl absorption (see discussion). C. Concomitant Rb absorption.
3-gram portions of roots were incubated separately for different time intervals in two or five litres of a solution containing 0.05 mM Tl_2SO_4, 0.1 mM $Ca(HCO_3)_2$, 1 mM $CaBr_2$, and different concentrations of Rb_2SO_4 as indicated; the pH was about 7. Root analysis was byfla mephotometry. The experiment was run in triplicate.

fig. 5A, a stimulation of Tl absorption by Rb after 3 hours is indicated most conspicuously at low Tl/Rb ratios.

Summarizing the foregoing, it may be stated that a strong mutual interference does indeed exist in Tl and Rb absorption. The extent to which the features of this interference comply with the pattern of competitive inhibition will be enlarged upon in the discussion.

Introduction of increasing amounts of Na into the experimental solution in-

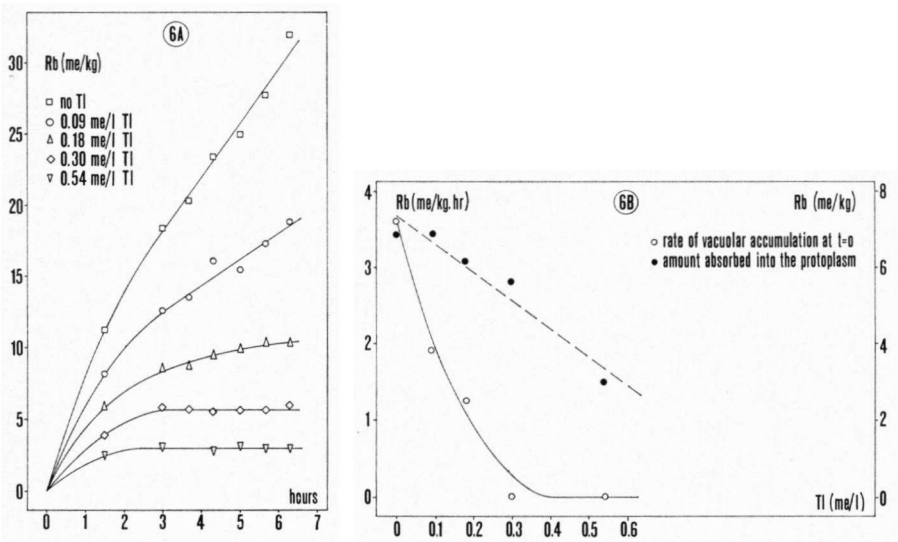
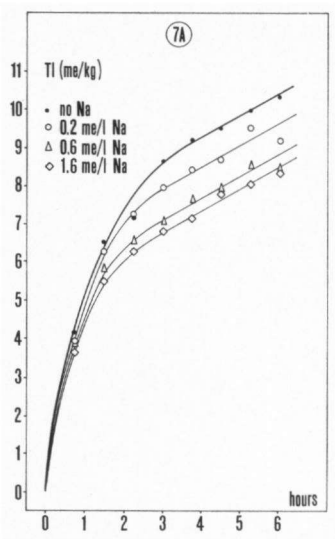
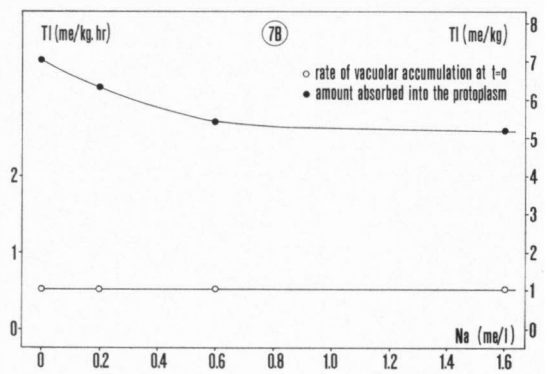


Fig. 6, A, B, and C. The effect of increasing Tl concentrations on Rb absorption from a 0.1 mM Rb_2SO_4 solution. A. Rb absorption. B. The separate effect on both components of Rb absorption (see discussion). C. Concomitant Tl absorption.
2-gram portions of roots were incubated separately for different time intervals in two litres of a solution containing 0.1 mM Rb_2SO_4, 0.1 mM $Ca(HCO_3)_2$, 1 mM $CaBr_2$, and different concentrations of Tl_2SO_4 as indicated; the pH was about 7. Root analysis was by flame-photometry. No replicate of the experiment was run.

fluences Tl absorption in a way that is clearly distinct from the effect of Rb, as is evident from a comparison of *figs. 7A* and *5A*. The absorption of Tl after 3 hours seems to be not at all affected by the presence of Na. On the other hand, the reduction in the rapid initial phase is slight and limited, an increase of the Na concentration from 0.6 to 1.6 me/l having hardly any further effect. Concomitant Na absorption is strongly repressed at 0.2 me/l (*fig. 7C; cf. fig. 3*) but even at 1.6 me/l, where Na absorption is larger, there is no stimulation of Tl absorption after 3 hours as there was in the case of Rb.





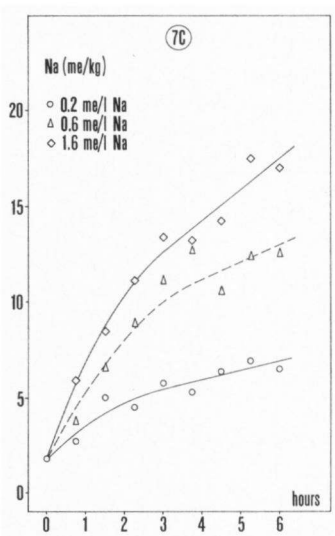


Fig. 7, A, B, and C. The effect of increasing Na concentrations on Tl absorption from a 0.1 mM Tl_2SO_4 solution. A. Tl absorption. B. The separate effect on both components of Tl absorption (see discussion). C. Concomitant Na absorption.
2-gram portions of roots were incubated separately for different time intervals in 2 litres of a solution containing 0.1 mM Tl_2SO_4 and 0, 0.1, 0.3 or 0.8 mM Na_2SO_4; the pH was about 6.1. Root analysis was by flame-photometry. No replicate of the experiment was run.

4. DISCUSSION

A crucial question with respect to the general shape of the relation between Tl absorption and time (*figs 1, A* and *B, 5A, 7A*) is whether a poisoning of general cell metabolism, as reflected in the gradual decline of the rate of respiration (*fig. 2*), must be held responsible for the low uptake values observed after about 3 hours. In the view of the authors, the experiments with Na (*figs. 3, A* and *B*) offer strong evidence against such an interpretation. The mutual interference

observed on the one hand in Rb and Tl absorption (see preceding chapter) and on the other in K (Rb) and Na uptake (BANGE *et al.* 1965; BANGE & HOOYMANS 1967) points to a close relationship between these absorption processes and therefore seems to justify the use of Na as a control for the effect of Tl on the absorption of the monovalent cations and thus on its own absorption.

Otherwise, the time curve of Tl uptake, like that of K (Rb) and Na absorption (HOOYMANS 1964), may be biphasic. This behaviour has been explained (HOOYMANS) by the assumption that in the absence of Ca during the first 2 to 3 hours, a direct uptake of the ion to the protoplasm is superimposed on steady-state vacuolar accumulation by a carrier mechanism. The effect of excess Ca already mentioned in the preceding section, would then consist of elimination of this direct pathway to the protoplasm, vacuolar absorption being insensitive to the divalent ion. When, in the presence of excess Ca, the rapid initial phase still persists, as in the case of Rb, at concentrations higher than about 0.03 me/l, the protoplasm is assumed to be able to withdraw ions from the carrier involved in vacuolar filling. The close agreement between the response of Tl uptake to excess Ca (*fig. 4*) with the behaviour of Rb in the concentration range from 0 to 0.2 me/l suggests that Tl uptake has a biphasic nature similar to that of Rb or Na absorption.

Additional support for this view is given by the competition experiments with Rb. The difference in the response of Tl uptake to Rb before and after 3 hours (*fig. 5A*) can be understood only if two components are involved in Tl absorption. It is this difference in response that sometimes yields a distinctly biphasic time curve, as for instance in *fig. 6C* at a Tl concentration of 0.09 me/l.

Thus, in Tl uptake both a protoplasmatic and a vacuolar component apparently are involved. The only feature distinguishing the absorption of Tl from that of the other monovalent cations is that vacuolar accumulation does not show a substantial steady-state but remains low and tends to stagnate, though this tendency is not always equally pronounced (*cf. figs. 1A* and *7A*). Therefore Tl seems to block the system that mediates its transport to the vacuole. This question will be discussed further below.

Additional evidence for a close correspondence between Tl uptake and the absorption of the other monovalent cations is furnished by the similar dependence on external concentration. It has been shown previously for Rb and Cs (BANGE & MEIJER 1966) that both uptake to the protoplasm and vacuolar accumulation show saturation kinetics. Tl uptake for 1 and 2 hours, which as discussed above represents mainly uptake to the protoplasm, behaves correspondingly.

All these facts suggest that Tl absorption is not an isolated phenomenon but that sites are involved that normally bind one of the monovalent nutrient cations. Three arguments favour the view that Tl takes the place of K and not of Na.

In the first place, half-saturation of the uptake is obtained at a concentration of about 0.005 me/l. Under similar conditions for Rb (unpublished results) the half-value of both the uptake to the protoplasm and the accumulation in the vacuole is of the same order of magnitude (around 5×10^{-6} M). In older work

this value was also found for K uptake by excised barley roots (BANGE *et al.* 1965). In contrast to these values, Na absorption shows half-saturation at concentrations about 10 times higher.

In the second place, as has already been pointed out, the response of Tl uptake to excess Ca is strongly reminiscent of the features of Rb absorption. So, as for Rb, in the presence of excess Ca the protoplasmatic sites can be saturated with Tl through the carrier system at relatively low external Tl concentrations. In the case of Na much higher Na concentrations are required to overcome the effect of Ca on the first phase of absorption (*cf.* HOOYMANS 1964).

The third argument can be found in the results of the competition experiments. *Figs 5B* and *6B* represent the influence of increasing concentrations of Rb and Tl, respectively, on either component of Tl and Rb uptake, respectively. Separation of the protoplasmatic and vacuolar components was achieved by determining the ordinate intercepts of the uptake curves of *figs 5A* and *6A*, respectively. For cases showing a decline in the uptake rate after 3 hours, this separation was performed tentatively on the assumption that there was a linear decline in the rate of vacuolar absorption during the whole experimental period. For the rate of vacuolar accumulation, the extrapolated value at $t = 0$ was used. From *fig. 5B* it appears that protoplasmatic Tl absorption is reduced to 50% at a Rb/Tl concentration ratio of 0.58 and to about 5% at a ratio of 2.5. In the opposite case (*fig. 6B*), 50% reduction of protoplasmatic Rb uptake is obtained at a Rb/Tl concentration ratio of 0.40, and, though complete suppression was not realized within the concentration ratios used, at the highest Tl concentration a considerable further decline of protoplasmatic Rb uptake is indicated.

The results therefore suggest that Tl and Rb can completely expel each other from the protoplasmatic sites as a competitive inhibition requires, the (apparent) affinity of Rb for these sites being about twice as large as that of Tl. The pattern of mutual interference in vacuolar accumulation is complicated by the facts mentioned in the preceding section. So the optimum curve in *fig. 5B* may be regarded as the result of the opposite effects of a stimulation predominating at low Rb concentrations and a competition getting the upper hand at higher Rb concentrations. On the other hand, in the curve for vacuolar Rb accumulation in *fig. 6B* an inhibiting effect of Tl on the release of Rb into the vacuole, which is the seamy side of the stimulating effect of Rb on Tl accumulation, may be superimposed on normal competition. The complex nature of these curves makes the quantitative aspects of the competition proper inaccessible.

The features of the mutual interference between Rb and Tl in both absorption components thus seem to be reconcilable with a competitive inhibition. This implies that the mutual interference between Tl and Na absorption must behave differently. It has been concluded from previous work (BANGE *et al.* 1965; BANGE & HOOYMANS 1967) that in the range of low concentrations K (Rb) and Na occupy separate absorption sites. Although Na uptake into the protoplasm as well as into the vacuole appears to be low in the presence of Tl (*fig. 7C*), a concomitant effect of Na on Tl absorption is small and restricted to part (about 20%) of the protoplasmatic uptake (*fig. 7, A* and *B*).

Consequently, the nature of the mutual interference together with the quantitative aspects of the saturation kinetics and of the Ca effect strongly suggest that Tl and Rb make use of the same sites that normally bind K. At the same time, as we have seen, the behaviour of Tl in the absorption process deviates considerably from that of K and Rb. Actually, these divergent features will be made use of to speculate about the mechanism of K absorption.

The process by which ions are released to the vacuole appears to be much more discriminative with respect to the nature of the ion than the process of binding to protoplasmatic or carrier sites. Thus, K, Rb, and Tl saturate the protoplasmatic sites either directly or through the carrier system at an equal rate and to a similar extent, whereas vacuolar release decreases in the sequence mentioned. When in vacuolar release an enzymatic breakdown of a chemical or structural nature of the ion-carrier complex is involved, the rate of this reaction is apparently dependent on the properties of the ion bound. This seems to imply that there is a structural interaction between the ion and the carrier molecule such that the binding of physiological isotopes leads to smaller or greater modifications in carrier structure and thus in rate of carrier breakdown. The interesting observation that the rate of Tl transport to the vacuole can benefit from the presence of Rb then finds its simplest interpretation as a relief of the strong deterioration of carrier structure brought about by Tl. Na does not bring about such relief, which is in accordance with the conclusions (BANGE *et al.* 1965) that Na and K are transported at separate sites. An obvious condition for this hypothesis is that a carrier molecule must have more than one site binding K.

A feature still requiring an explanation is the more or less pronounced and steady slowing down of vacuolar Tl absorption, a phenomenon also shown by concomitant Rb absorption at relatively high Tl/Rb ratios but likewise relieved by larger relative amounts of Rb (*figs. 5, A* and *C*) even to the degree of a completely steady vacuolar Tl and Rb accumulation (*fig. 6C*). When the relative ease with which Tl ions are released from the separate sites on a carrier molecule differs from one site to another, sites from which the release is most difficult might increase in number during functional carrier turnover. Former work on Rb/Cs interactions (BANGE & MEIJER 1966) has suggested some heterogeneity of sites, since it was observed that a small uptake of Cs into the tissue persists even at an excess of Rb and that the converse also holds.

The large difference in response shown by the structures involved in vacuolar K accumulation with respect to the nature of the cation transported (K, Rb, or Tl), must be based on differences in the physico-chemical properties of the ions concerned. Apparently, the close correspondence between the values of their hydrated or crystalline radii (*table 1*) enables them to occupy identical sites, but the small differences between these values show no correlation with the rates of vacuolar accumulation observed. However, there is a correlation with the polarizability, which suggests that this property is an important factor in the structural interactions between ion and carrier.

The interest of the experiments described, however, lies not only in the behaviour of Tl absorption *per se*. It is hoped that the density of the electron-cloud

Table 1. Crystalline and hydrated radii and polarizability of K, Rb, and Tl ions.

	Crystalline radius[1]	Hydrated radius[1]	Polarizability[2]
K	1.33 Å	3.31 Å	2.12 Å^3
Rb	1.48	3.29	3.57
Tl	1.44	3.30	8.5

[1] Data from NIGHTINGALE (1959)
[2] Data from PAULING (1927) as cited by COHEN (1962)

of the Tl atom may make it possible to localize the ion at the subcellular level by electron microscopy and thus to use it as a 'tracer' for K. This will be the object of future work.

ACKNOWLEDGEMENTS

The technical assistance of Miss P. M. van der Duyn Schouten is gratefully acknowledged.

REFERENCES

BANGE, G. G. J. & J. J. M. HOOYMANS (1967): Transport of monovalent cations into, through and out of barley roots: an experimental theory. *Isotopes in Plant Nutrition and Physiology*: 249–263. International Atomic Energy Agency, Vienna.

— & C. L. C. MEIJER (1966): The alkali cation carrier of barley roots: a macromolecular structure? *Acta Bot. Neerl.* **15**: 434–450.

—, J. TROMP & S. HENKES (1965): Interactions in the absorption of potassium, sodium and ammonium ions in excised barley roots. *Acta Bot. Neerl.* **14**: 116–130.

BRITTEN, J. S. & N. BLANK (1968): Thallium activation of the (Na^+–K^+)-activated ATP-ase of rabbit kidney. *Biochim. Biophys. Acta* **159**: 160–166.

COHEN, D. (1962): Specific binding of rubidium in Chlorella. *J. Gen. Physiol.* **45**: 959–977.

HOOYMANS, J. J. M. (1964): The role of calcium in the absorption of anions and cations by excised barley roots. *Acta Bot. Neerl.* **13**: 507–540.

NIGHTINGALE, E. R. (1959): Phenomenological theory of ion solution. Effective radii of hydrated ions. *J. Physic. Chem.* **63**: 1381–1387.