THE ROLE OF CALCIUM IN SELECTIVE CATION UPTAKE
BY PLANT ROOTS

II. Effects of Temperature, Desorption Treatment
and Sodium Salt on Rubidium Uptake*

Toshio KAWASAKI and Shiro HORI**

In the previous paper (1), the effect of calcium on the selective uptake of monovalent cations was studied in relation to monovalent and divalent cation concentrations in the absorption solution. Data presented in that paper showed that the uptake of rubidium by plant roots was stimulated in the presence of calcium at a relatively high concentration of monovalent cations in the absorption solution. However, the uptake of sodium was inhibited by calcium at both the low and high concentration of monovalent cations. Further, it was suggested that the site of stimulation of calcium on rubidium uptake might be located in the metabolic process of ion uptake.

The present investigation was undertaken to extend the results of the previous paper, and to study in more detail the mechanism(s) of the calcium effect on the selective uptake of monovalent cations by plant roots. For this purpose, the calcium effect was examined at different experimental temperatures, with and without the desorption treatment of roots, and in the presence and absence of sodium in the absorption solution.

MATERIALS AND METHODS

Preparation of Root Materials

Excised roots of barley (Hordeum vulgare L., variety Akashinriki) were used. Root materials were prepared according to the procedure described previously (1).

Experimental Procedure

To investigate the effect of calcium or magnesium on the uptake of rubidium and sodium, equimolar mixtures of rubidium chloride and sodium chloride were used as the absorption solution. In experiments on the effect of sodium on rubidium uptake, absorption solutions consisted of a constant concentration of rubidium chloride and varying


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concentrations of sodium chloride. Absorption solutions were labeled with radioactive rubidium (^86Rb) or radioactive sodium (^22Na). The initial pH of the absorption solutions was adjusted to 5.4–5.7.

The procedures for the absorption experiment and the washing operation for roots were virtually identical with those described in the previous paper (1). In all experiments, 0.5 g of excised roots were used in 500 ml of absorption solution. The absorption period was 60 minutes, during which the solutions were continuously aerated and kept at 25±0.1°C or 2±0.5°C.

At the end of an absorption period, the roots were separated from the solution. Then the roots were blotted with filter paper and shaken for 30 seconds in 30 ml of demineralized water. This washing operation was repeated once more.

Roots of certain plots underwent desorption treatment, i.e., after washing the roots were shaken repeatedly for 30 minutes in 50 ml of desorption solution, then were blotted with filter paper. Desorption solutions used in the experiment on the effect of divalent cations on the uptake of monovalent cations had the same composition of monovalent cations as the absorption solution. In the experiment on the effect of sodium on the uptake of rubidium, a rubidium chloride solution (1.0 mM) was used as the desorption solution. All desorption solutions were non-radioactive, and no divalent cations were added to them.

**Radioactive Assay**

The procedure for the radioactive assay was identical with that described in the previous paper (1). Root samples were dried and ignited. One ml of dilute nitric acid solution was added to the ash and the samples were dried. Then, the samples were counted with a Geiger-Müller counter.

**RESULTS**

**Influences of Experimental Temperature and Desorption Treatment on the Calcium Effect**

Effects of divalent cations on the uptake of rubidium and sodium by excised barley roots from equimolar mixtures of rubidium chloride and sodium chloride were investigated under various conditions, with and without the desorption treatment of roots after an absorption period at 25°C and 2°C.

Figs. 1 and 2, respectively, show the rates of rubidium uptake from absorption solutions containing 0.001 and 1.0 mM of monovalent cations. Numbers on bars indicate the ratios of the results when calcium chloride or magnesium chloride was added to that when there was no addition
Fig. 1. Effects of divalent cations on the rubidium uptake with or without the desorption treatment. (Rubidium chloride and sodium chloride in absorption solution: 0.001 mM)

Fig. 2. Effects of divalent cations on the rubidium uptake with or without the desorption treatment. (Rubidium chloride and sodium chloride in absorption solution: 1.0 mM)

of divalent cations. The same system is used in the figures described below, except for Figs. 5 and 6. At the lower concentration (0.001 mM) of monovalent cations in the absorption solution, the rubidium uptake decreased in the presence of calcium or magnesium chloride, with or without the desorption treatment at 25°C and 2°C. At the relatively high concentration (1.0 mM) of monovalent cations, the rubidium uptake increased in the presence of calcium and magnesium chlorides both with and without the desorption treatment at 25°C. At 2°C, however, the rubidium uptake decreased in the presence of calcium and magnesium chlorides when there was no desorption treatment, whereas it increased with desorption treatment. Further, the stimulating effect of calcium chloride on rubidium uptake was larger than that of magnesium chloride.
The rates of sodium uptake from absorption solutions containing 0.001 and 1.0 mM of monovalent cations are shown in Figs. 3 and 4. 

Fig. 3. Effects of divalent cations on the sodium uptake with or without the desorption treatment. (Rubidium chloride and sodium chloride in absorption solution: 0.001 mM)

Fig. 4. Effects of divalent cations on the sodium uptake with or without the desorption treatment. (Rubidium chloride and sodium chloride in absorption solution: 1.0 mM)

respectively. Sodium uptake decreased in the presence of calcium and magnesium chlorides under all the conditions of the experiments; at the low and high concentrations of monovalent cations in the absorption solutions, with or without the desorption treatment of roots after the absorption period, and at both 25° and 2°C.

*Influence of Sodium Salt on Rubidium Uptake in Relation to the Presence and Absence of Calcium*

The effects of increasing concentrations of sodium chloride on rubidium uptake from a constant concentration of rubidium chloride solution were examined in the absence of calcium.
Fig. 5 shows the rates of rubidium uptake from an absorption solution which contained 0.001 mM of rubidium chloride and 0-1.0 mM of sodium chloride, at 25° and 2°C. Fig. 6 shows the rates of rubidium uptake from an absorption solution which contained 0.001 mM of rubidium chloride and 0-10.0 mM of sodium chloride. Numbers on bars indicate the ratios of the results when varying concentrations of sodium chloride were added to the results of no addition. At both the low (0.001 mM) and relatively high (1.0 mM) concentrations of rubidium chloride, the rubidium uptake decreased with increasing concentrations of sodium chloride in the absorption solution, with or without the desorption treatment at 25° and 2°C.

Subsequently, in order to determine the role of sodium in the calcium effect, a comparison was made between the rubidium uptake from a single salt solution of rubidium chloride (1.0 mM) and from an equimolar
mixture of rubidium and sodium chloride (1.0 mM each) in the presence and absence of calcium.

Figs. 7～9 show data from the experiments at various temperatures. These results showed that even when the temperature during the absorption period was 25°C, the calcium effect on the rubidium uptake from a single salt solution of rubidium chloride was nil or very small both with and without the desorption treatment. However, the calcium effect
on the rubidium uptake from a mixture of rubidium and sodium chlorides was clear. Table 1 summarizes the data shown in Figs. 7~9 based

![Figure 9](image.png)

**Fig. 9.** Rubidium uptake from rubidium chloride and rubidium plus sodium chloride solutions in the presence and absence of calcium. (Rubidium chloride or sodium chloride in absorption solution: 1.0 mM; Temperature: absorption 25°C, washing 25°C, desorption 25°C)

<table>
<thead>
<tr>
<th>Composition of absorption solution</th>
<th>Rubidium uptake (%)</th>
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<tbody>
<tr>
<td>Rb</td>
<td>100</td>
</tr>
<tr>
<td>Rb + Na</td>
<td>66</td>
</tr>
<tr>
<td>Non-desorption</td>
<td></td>
</tr>
<tr>
<td>Rb</td>
<td>100</td>
</tr>
<tr>
<td>Rb + Na</td>
<td>89</td>
</tr>
<tr>
<td>Desorption</td>
<td></td>
</tr>
<tr>
<td>Rb</td>
<td>100</td>
</tr>
<tr>
<td>Rb + Na</td>
<td>101</td>
</tr>
</tbody>
</table>

**TABLE 1**

Repressive effect of sodium on rubidium uptake in the presence and absence of calcium

<table>
<thead>
<tr>
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<tbody>
<tr>
<td></td>
<td>Rb</td>
<td>Rb + Na</td>
<td>Rb</td>
</tr>
<tr>
<td>Washing</td>
<td>Rb</td>
<td>Rb + Na</td>
<td>Rb</td>
</tr>
<tr>
<td>Desorption</td>
<td>Rb</td>
<td>Rb + Na</td>
<td>Rb</td>
</tr>
<tr>
<td>Non-desorption +Ca</td>
<td>100</td>
<td>66</td>
<td>100</td>
</tr>
<tr>
<td>+Ca</td>
<td>100</td>
<td>87</td>
<td>100</td>
</tr>
<tr>
<td>Desorption  +Ca</td>
<td>100</td>
<td>63</td>
<td>100</td>
</tr>
<tr>
<td>+Ca</td>
<td>100</td>
<td>87</td>
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on the ratio of the rubidium uptake from a rubidium plus sodium chloride solution to that from a single salt solution of rubidium chloride. Under all the experimental conditions, the rubidium uptake decreased on the addition of sodium to the absorption solution. However, the repressive effect of sodium on the rubidium uptake was less when calcium was present in the absorption solution than when it was absent.
DISCUSSION

Before discussing the above data, we should consider the characteristics of the exchangeable and non-exchangeable fractions of ions in plant roots. Some studies have been done on the passive and active processes of ion uptake by plant roots (2, 3). From the results, it has generally been accepted that ions gained by the active process of ion uptake are largely non-exchangeable with ambient ions of the same or other species (4). Thus the non-exchangeable fraction of ions taken up by plant roots must be related to the active (metabolic) process of ion uptake, and the exchangeable fraction to the passive (non-metabolic) process. In the present paper, the ions retained in the roots after the desorption treatment correspond to the non-exchangeable fraction, and the ions taken up in the roots without the desorption treatment after washing correspond to the sum of the non-exchangeable and exchangeable fractions.

It has been reported that calcium increased the affinity between potassium (or rubidium) ion and its carrier, based on the ion-carrier complex theory (5–7). In the results of this investigation also, the stimulation of rubidium uptake by divalent cations (especially the calcium ion) was larger with the desorption treatment than without the desorption treatment (Fig. 2). Since the non-exchangeable fraction of ions taken up by plant roots is closely related to the active (metabolic) process of ion uptake, the stimulating effect of calcium on the rubidium uptake is thought to be dependent on the active (metabolic) process of ion uptake. Furthermore, since the metabolic activity of living cells decreases at low temperatures, the fact that the calcium effect on rubidium uptake was reduced or abolished by low temperature treatment (2°C) supports the above hypothesis (Fig. 2).

On the other hand, it was found that the calcium effect on the rubidium uptake from a rubidium plus sodium chloride solution was greater than that from a single salt solution of rubidium chloride (Figs. 7–9). These results suggest that sodium plays an important role in the mechanism(s) of the calcium effect on rubidium uptake. Although the rubidium uptake decreased on the addition of sodium, whether in the presence or absence of calcium (Figs. 7–9), this repression of the rubidium uptake by sodium was smaller in the presence of calcium than in its absence (Table 1). Jacobson et al. (8, 9) and Waisel (10) suggested that the stimulating effect of calcium on potassium or rubidium uptake was due essentially to blockage of interfering ions.

Though the uptake of potassium or rubidium was reduced by the addition of sodium (11), both carriers (or carrier sites) for potassium or rubidium were posited, which were difficultly and easily combined by sodium (12–14). In this paper, the rubidium uptake decreased with
Increasing concentrations of sodium chloride, but the inhibition by sodium was not equivalent to rubidium. In addition, some reports showed that the sodium uptake decreased markedly in the presence of calcium (1, 9, 10, 14-16). Accordingly, it is reasonable to consider that, as a cause of the stimulation of rubidium uptake by calcium, calcium reversed the repression of rubidium uptake by sodium.

**SUMMARY**

Effects of calcium on the selective absorption of monovalent cations were studied under various conditions. The role of sodium in the stimulating effect of calcium on rubidium uptake was also investigated.

1) At a low concentration (0.001 mM) of monovalent cations in an equimolar mixture of rubidium chloride and sodium chloride, the rubidium and sodium uptake decreased in the presence of calcium or magnesium chloride, both with and without the desorption treatment of plant roots at 25°C and 2°C.

2) At a higher concentration (1.0 mM) of monovalent cations, the sodium uptake decreased at 25°C and 2°C, and the rubidium uptake increased at 25°C in the presence of calcium or magnesium chloride with or without the desorption treatment. However, at 2°C, calcium and magnesium chlorides decreased the rubidium uptake without the desorption treatment of plant roots, but increased it with the desorption treatment.

Stimulation of the rubidium uptake was greater in the presence of calcium than in the presence of magnesium.

3) The stimulating effect of calcium was more marked when sodium was present in the absorption solution.

4) It has been posited here, as noted in the previous paper, that the calcium effect on the rubidium uptake was dependent on the metabolic process of ion uptake. Moreover, the results support the contention that sodium plays an important role in the mechanism(s) of calcium effect.

**REFERENCES**

