Short Communication

Influence of the Root Tip and the Duration of Washing on \( K^+ \) Retention by Excised Apical Root Segments of Corn

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ABSTRACT

Excised apical segments of corn root (Zea mays) (5-15 mm from the root cap), some with and some without the root tips (0-5 mm) attached, were washed for varying time periods up to 4 hours in 0.5 mM CaSO_4. After washing, tips were removed from those segments washed with tips attached, and then all segments and tips were analyzed for \( K^+ \) content. The root tips (0-5 mm) initially contained about twice the \( K^+ \) of the apical segments (5-15 mm). The loss of \( K^+ \) did not exceed 15% in the tips or 20% in the apical segments. Loss of \( K^+ \) was most pronounced during the first hour of washing. There was little difference in \( K^+ \) content of apical segments washed with tips attached compared with those washed tipless. Thus, the presence of the intact root tip had no consistent influence on the ability of the older root tissue to retain \( K^+ \).

Numerous studies in the last 10 years have shown that "washing" of excised root tissue in water or aqueous solutions subsequently results in enhanced rates of mineral ion accumulation (4). The physiological basis for this response, however, has not yet been identified.

Leonard and Hanson (2), working with excised corn roots, showed that the enhancement of mineral ion accumulation increased with the length of the incubation or washing period. Parra and Smith (3) found that enhancement obtained when the root tip was left attached during the washing period was less than one-half that obtained when the tip was removed prior to the washing period. This led them to conclude that the enhancement is caused by the leaching from the tissue of some substance which inhibits ion accumulation and which originates in the root tip.

When excised root tissue is exposed to the conditions of washing, some soluble ions are lost from the tissue, and it has been suggested that this loss is the basis for the washing response. The explanation is that tissue which has lost part of its ionic content will subsequently absorb ions at a faster rate than tissue with which had not lost these ions. However, Leonard and Hanson (2) found that whereas the loss of \( K^+ \) occurred at both cold and warm temperatures, enhancement of \( K^+ \) absorption occurred only at warm temperatures. Their interpretation of this was that enhancement of absorption rates could not be attributed to the loss of salt during washing. The present paper supports their conclusion on an entirely different experimental basis—here the purpose was to determine whether the presence of the attached root tip, which is known to influence enhancement, could also influence the degree to which \( K^+ \) is retained by other parts of the root.

MATERIALS AND METHODS

Seeds of corn (Zea mays, DeKalb XL 80) were surface-sterilized with Clorox, then soaked in a flask of glass-distilled H_2O that was changed every 8 hr and given vigorous aeration. After 24 hr, the germinating seeds were planted on cheesecloth laid over a stainless steel mesh which was suspended at the top of an 8-liter polyethylene container filled with 0.2 mM CaSO_4 solution. The seedlings were grown in a dark incubator at 28 C and given constant aeration. Every 24 hr, the seedlings were rinsed and the CaSO_4 solution replaced with fresh solution.

After 4 days, segments were excised from the primary roots. Half of the samples consisted of segments taken 0 to 15 mm from the root apex. The rest were segments excised from 5 to 15 mm from the apex and the tips (0-5 mm) discarded. Each sample, consisting of 15 root segments, was placed in a Fiberglas screen bag and immersed in a separate 4-liter beaker of 0.5 mM CaSO_4 washing solution. This solution was given constant aeration and held at 30 C. Unwashed samples were prepared as controls. Pairs of experimental samples—one containing segments with tips still attached and one without—were immersed in the washing solutions for 1, 2, or 4 hr.

After washing, the tips (0-5 mm) were removed from those segments with tips still attached. At this time, all segments were blotted dry, weighed, and the samples placed individually in 50-ml beakers. The excised tips were treated in the same manner. All samples were ashed at 520 C for 45 min. The ashes were digested in a small volume of hot concentrated HNO_3 which was then evaporated to dryness. Finally, the digested residues were redissolved in distilled H_2O and analyzed for \( K^+ \) content using flame emission spectroscopy.

The above procedure was carried out four times, yielding results which showed only small quantitative differences.

RESULTS AND DISCUSSION

The initial \( K^+ \) content of the tips (0-5 mm) was about twice that of the apical segments (5-15 mm), a difference consistent with earlier findings (1). All root segments lost a small amount of \( K^+ \) during the washing period (Table 1). There was no possibility of confusion of \( K^+ \) loss or reabsorption between samples during the washing period because each sample was washed separately from all other samples. Furthermore, the volume of the washing solution (4000 ml) was very large compared to the size of each sample (maximum weight 90 mg), so that the time course of loss should not have been influenced by appreciable changes in external \( K^+ \) concentration.

In apical segments washed without their tips, the \( K^+ \) content dropped during the first hour of washing to about 86% of the initial value. Samples washed for longer periods (4 hr) consist-
TABLE I. Changes in the potassium content of corn root segments washed for varying time periods.

Excised corn root segments were washed with or without attached tips for varying time periods. At the end of the washing period tips were removed from those segments washed with attached tips, and then all segments and tips were analyzed for K+ content, which is presented as μmoles K+ / g fresh wt. Values are averages of four experimental determinations of 15 segments each.

<table>
<thead>
<tr>
<th>Duration of washing period, hr</th>
<th>0</th>
<th>1</th>
<th>2</th>
<th>4</th>
</tr>
</thead>
<tbody>
<tr>
<td>Tips</td>
<td>81.6 ± 9.7</td>
<td>74.6 ± 14.8</td>
<td>75.1 ± 14.0</td>
<td>73.0 ± 13.0</td>
</tr>
<tr>
<td>Apical segments, tips attached</td>
<td>38.5 ± 1.3</td>
<td>34.6 ± 4.7</td>
<td>34.6 ± 3.7</td>
<td>35.0 ± 4.2</td>
</tr>
<tr>
<td>Apical segments, tips removed</td>
<td>38.5 ± 1.3</td>
<td>33.2 ± 4.9</td>
<td>35.5 ± 5.6</td>
<td>37.4 ± 4.2</td>
</tr>
</tbody>
</table>

ently had K+ contents slightly higher than those washed for only 1 hr. This is interpreted to indicate that samples washed without their tips underwent the typical washing response (3), and therefore, began to reaccumulate part of the K+ lost initially. In apical segments washed with tips intact and attached, the loss of K+ showed no significant change after the initial drop to 90% of the initial value during the 1st hr. The tips (0-5 mm) lost about the same proportional amount of their K+ as did the apical segments to which they were attached during the washing period.

Among the repetitions of this experiment, neither of the two groups of apical segments consistently maintained a K+ content higher or lower than the other except at the 4-hr sampling when the segments washed tipless always had a slightly higher K+ content. This general similarity is in striking contrast to the large and consistent difference in ion-absorption rates found between the two groups (3) which persists at least 6 hr. Segments washed without their tips absorbed Rb+ at least twice as rapidly as those washed with tips intact. If the enhancement were caused by loss of K+ during washing, then segments with essentially identical K+ contents should not show the response. This does not correspond with the data presented, and therefore, we concluded that the enhancement response is not due to loss of K+ during the washing period.

LITERATURE CITED