Inhibition of Ca^{2+} and Zn^{2+} Uptake by Mn^{2+} in Excised Rice Roots

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Ion Inhibition

The effects of varying concentrations of Mn^{2+} on the absorption of divalent cations, *viz.*, Ca^{2+} and Zn^{2+} from 2 concentrations were studied. Mn^{2+} at all concentrations tested were inhibitory to Ca^{2+} uptake from 0.1 and 5 mM $CaCl_2$. However, when Ca^{2+} uptake was measured at concentrations ranging from 0.05 to 10 mM $CaCl_2$, $MnCl_2$ only at high concentration of 5 mM, inhibited Ca absorption. Zn^{2+} uptake from 0.1 mM $ZnCl_2$ was decreased by all concentrations of Mn^{2+}.

One of the most intriguing problems in plant nutrition is the availability of micronutrients, especially Fe^{2+} and Mn^{2+}, which are poorly absorbed by, and less translocated from the roots to the shoot. Our studies in this respect have revealed several important findings [1–5], and the interaction between the absorption of these elements appears to play a significant role in their mutual availabilities.

It has been known that elements which are chemically related, mutually interact in their absorption. Although the ion-carriers are considered to be specific to the individual ions, competition for the carrier-sites by similar as well as dissimilar ions is also recorded [6]. The manner in which this competition takes place is still a matter of conjecture. Eisenman [7] has indicated a number of sequences for competition between monovalent cations like Cs^+, Rb^+, K^+ and Na^+.

From our earlier studies on the mechanisms of absorption and transport of Fe^{2+} and Mn^{2+}, and also on the effects of other cations on the absorption of Mn^{2+} by excised rice roots [3, 4], it was anticipated that low and high concentrations of Mn^{2+} would also affect the uptake of other cations. The presence of high amounts of Mn^{2+} is toxic to plants and the precise nature of this toxicity is however not known. It has recently been shown that Mn^{2+} interferes with Fe^{2+} utilisation in chlorophyll synthesis [8]. The present report deals with the effects of Mn^{2+} on the absorption of Ca^{2+} and Zn^{2+}, by varying not only the concentrations of Ca^{2+} and Zn^{2+} but also that of Mn^{2+}. The uptake of ions by excised roots reflects the uptake mechanisms at cellular level, and the low and high concentrations of ions employed here reflect those of dual mechanisms of Epstein [6], who has described that mechanism 1 is functional in the low ion concentrations from 0.1 to 0.5 mM, and mechanism 2 operates in the high ion concentrations above 1 to 50 mM. Earlier [4], we reported the effects of Ca^{2+}, Fe^{2+}, Mg^{2+}, K^+, and Na^+. We have studied also the action of Mn^{2+} on the absorption of monovalent cations, *viz.*, Na^+, K^+, and Rb^+ by excised rice roots [9]. The present report is complementary to our earlier investigations, and is limited to 2 divalent cations, *viz.*, Ca^{2+} and Zn^{2+}, leaving Mg^{2+} and Cu^{2+} for future studies. The mechanisms of ion absorption have been discussed recently [10], and the objective of our study is to understand the interactions, and not concerned with the mechanisms of ion uptake *per se*.

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Materials and Methods

The procedures for growing and obtaining rice (*Oryza sativa* L. cv. I.R. 8) roots and experimentation are those as described earlier [3, 4]. The rates of absorption were measured by suspending the excised roots through nylon-net bags into the labelled solutions of $CaCl_2$ and $ZnCl_2$, in the absence and presence of different concentrations of $MnCl_2$. The samples were desorbed for 15 min in cold (5 °C) unlabelled solutions of the respective salts, and then radioassayed. The pH of the experimental solutions was adjusted to 5.5. In all experiments excepting where Ca^{2+} was measured, 0.1 mM $CaSO_4$ was routinely added in the medium in order to maintain the membrane permeability properties. $^{45}Ca^{2+}$ and $^{65}Zn^{2+}$ were used for labelling $CaCl_2$ and $ZnCl_2$ solutions respectively. $^{45}Ca^{2+}$ was assayed in a liquid scintillation spectrometer and $^{65}Zn^{2+}$ was analysed in a gamma ray spectrometer. Standard errors of the means of triplicate values are shown by vertical bars in the figures.

Results and Discussion

Figure 1 illustrates the effects of different concentrations of Mn^{2+} on Ca^{2+} uptake from 0.1 and 5 mM

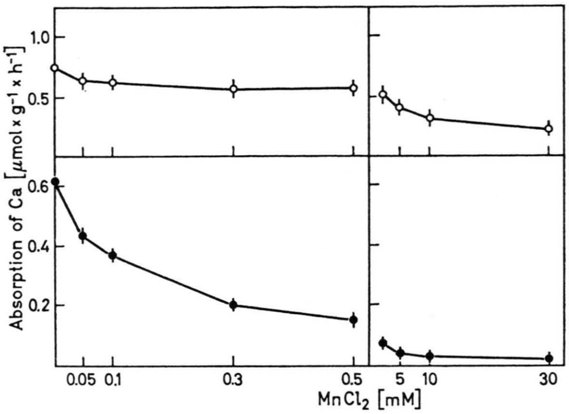

Fig. 1. The rates of absorption of Ca^{2+} from 0.1 and 5 mM $CaCl_2$, in the absence and presence of varying concentrations of $MnCl_2$. ○—○, 5 mM $CaCl_2$; ●—●, 0.1 mM $CaCl_2$.

$CaCl_2$. Although the absorption from both concentrations is decreased by Mn^{2+}, it is more drastic in the uptake from 0.1 mM. The absorption from 5 mM $CaCl_2$ is reduced by Mn^{2+} from 0.05 mM and above, although there is no significant further reduction above 0.05 mM. We have examined 0.1 and 5 mM $CaCl_2$, and these represent the mechanism 1 and mechanism 2 respectively as described by Epstein [6]. Although the soil solution contains widely varying concentrations of salts in general, and Ca^{2+} in particular, we are here interested in the uptake of these cations at cellular level, from a solution culture. It is observed that low concentrations of $MnCl_2$ upto 0.5 mM, reduced Ca^{2+} uptake from 5 mM $CaCl_2$ to nearly 20%, while there is greater reduction nearly 41% for the uptake from 0.1 mM $CaCl_2$ (Fig. 1). This effect is perhaps due to the concentrations, since $CaCl_2$ concentration of 5 mM is very high compared to $MnCl_2$, and the latter therefore is not able to compete with Ca^{2+}. This is supported by the observation that when $MnCl_2$ concentration is raised (1 to 30 mM), Ca^{2+} uptake is decreased.

In contrast to Fig. 1, the data in Fig. 2 describe the rates of Ca^{2+} uptake, varying the $CaCl_2$ concentration in the medium, and keeping only 2 concentrations of the interfering cation Mn^{2+} (0.1 and

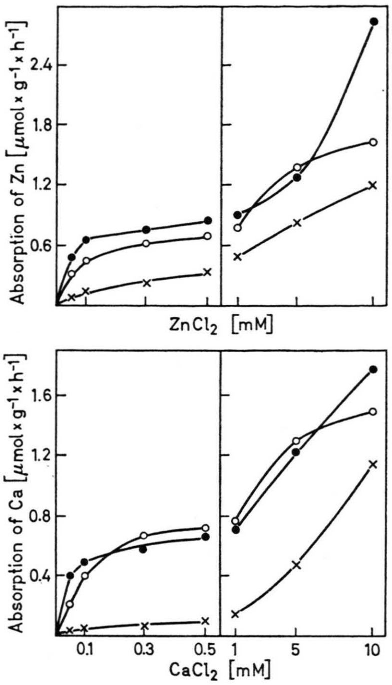

Fig. 2. The rates of absorption of Ca^{2+} and Zn^{2+} from different concentrations of $CaCl_2$ and $ZnCl_2$, in the absence and presence of 0.1 and 5 mM $MnCl_2$. ●—●, Control; ○—○, 0.1 mM $MnCl_2$; ×—×, 5.0 mM $MnCl_2$.

5.0 mM $MnCl_2$). It is seen that Mn^{2+} at 5 mM is greatly inhibitory to Ca^{2+} uptake. Earlier, we have observed that 0.5 mM Ca^{2+} inhibited Mn^{2+} absorption from low concentration range in a competitive manner [4]. In the light of the present findings, it is con-

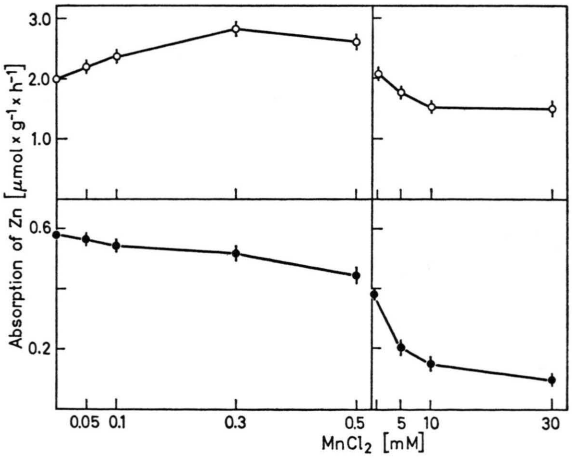

Fig. 3. The rates of absorption of Zn^{2+} from 0.1 and 5 mM $ZnCl_2$, in the absence and presence of varying concentrations of $MnCl_2$. ○—○, 5 mM $ZnCl_2$; ●—●, 0.1 mM $ZnCl_2$.

cluded that Ca^{2+} and Mn^{2+} are mutually inhibitory in their absorption, and perhaps these 2 cations have a regulatory effect on the absorption of elements which may otherwise prove toxic in high concentrations. Viets [11] had suggested that cations act directly on cell membranes and regulate ion absorption. A major role of Ca^{2+} in plant nutrition appears to be that of preventing the toxicities of heavy metals [12]. Since Ca^{2+} uptake is also inhibited by Mn^{2+}, Ca^{2+} perhaps renders this protective effect, by undergoing an inhibition of its own absorption.

Mn^{2+} in the concentration range of 1 to 30 mM is found to reduce Zn^{2+} uptake from 0.1 and 5 mM $ZnCl_2$ (Fig. 3). On the other hand, when Mn^{2+} concentration is low (0.05 to 0.5 mM), the uptake from 0.1 mM $ZnCl_2$ is slightly reduced, and that from 5 mM is enhanced. The promoting effect of Mn^{2+} on Zn^{2+} uptake differs from that on Ca^{2+} uptake from 5 mM (Fig. 1). No explanation for this difference can be offered at present. However, there is always a difference in the ion uptake behaviour with different cations, and what is true for Ca^{2+} need not be true for Zn^{2+}. It has been observed that low concentrations of K^+ or Mg^{2+} promoted Mn^{2+} uptake from high Mn^{2+} concentrations [4]. Results in Fig. 2 reveal that only high concentrations of Mn^{2+} (5 mM) is inhibitory to Zn^{2+} uptake from all concentration ranges. This is equally true for Ca^{2+} uptake also (Fig. 2). While the inhibition of Ca^{2+} by Mn^{2+} appears to be of a competitive nature because the inhibition increases with increasing concentrations of Mn^{2+} (Fig. 1), the inhibition of Zn^{2+} uptake is noncompetitive, and is similar to the inhibition of ions by polyvalent cations [13].

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