Effect of pH and iron complexes on the iron species found in the roots of iron deficient cucumber after iron supply

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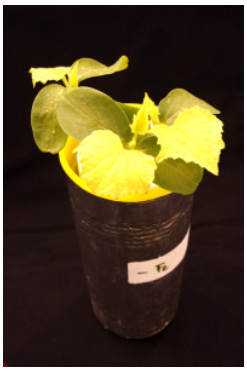


BACKGROUND

I. Introduction

Iron chelates has playing a key role in the remediation of iron deficiency in recent agricultural practices. They form stable complexes with iron but they persist without decomposition for a long time in natural ecosystems flowing in the soil water so the need for biodegradable alternatives emerged. Fe-citrate is a natural iron complex normally found in plants and its ability to provide iron for plants is comparable to other chelate as Fe-EDTA, Fe-EDDHA. However they differ in stability and the ability to deliver Fe to the sites of reduction and uptake [1].

Previous studies show that the pH of the nutrient solution strongly influence the adsorption, uptake and translocation of iron in plants. It may alter the stability of chelates and the activity of membrane bound enzymes involved in the iron acquisition of Strategy I plants [2].

II. Objectives

The aim of this study was to investigate the effect of pH and different Fe-complexes on the uptake and accumulation of iron in cucumber roots with the help of ^{57}Fe Mössbauer spectroscopy.

III. Experimental

Cucumber (Cucumis stivus L. Joker F1) plants were grown in modified quarter strength Hoagland nutrient solution without iron for two weeks. Then iron was supplied as Fe-citrate, Fe-EDTA or Fe-EDDHA in 0.5 mM concentration for 30 min and the roots were immediately frozen in liquid N_2. In a separate experiment Fe-citrate was supplied in different Fe:Cit ratios: 3.1, 1:1, 1:50, 1:100.

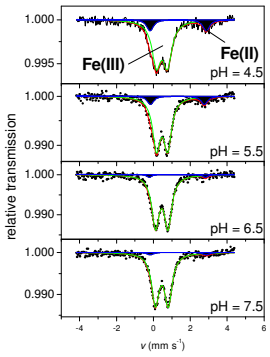
The effect of pH was measured as follows: 0.5 mM Fe-citrate (1:1,1) was supplied for 30 min in basic nutrient solution buffered to pH=4.5, 5.5, 6.5 (MES) and 7.5 (HEPES), respectively. Then the roots were immediately frozen in liquid N_2.

All frozen roots were measured by Mössbauer spectroscopy and also by ICP. Data of Fe concentration were also assessed from the Mössbauer spectra.

All Fe-complexes were prepared from ^{57}Fe enriched (95%) $^{57}FeCl_3$ in water solution. In the case of FeEDTA and Fe-EDDHA, the Fe:ligand ratio was 1.1:1. The pH of the Fe-solutions was adjusted to 5.5.

RESULTS AND DISCUSSION

IV. Effect of pH on the iron components found in the roots of iron deficient cucumber

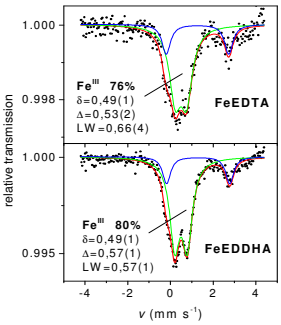


•According to the Mössbauer parameters, the Fe^{II} component can be assigned to Fe^{II}-hexaaqua complex (δ=1.34(2) mms^{-1} Δ=3.04(4) mms^{-1} LW=0.44(6) mms^{-1}). The relative amount of Fe^{II} decreases from 16% to 4 % as the pH is increased from 5.5 to 7.5.

• In the case of pH=4.5 and 5.5 the Fe^{III} component (δ=0.47(1) mms^{-1} Δ=0.60(1) mms^{-1} LW=0.55(2) mms^{-1}) can be identified as Fe^{III}-carboxylate complexes (e.g. Fe-Citrate) [3]. In the case of pH=6.5 and 7.5, the quadrupole splitting of the Fe^{III} component increases (Δ=0.70(1) mms^{-1}) which suggest the formation of hydrous ferric oxides-hydroxides and/or the change of the structure of Fe^{III}-carboxylate complexes.

• Both the total Fe and the total Fe^{II} concentrations are decreasing with increasing pH.

V. Effect of iron complexes on the iron components found in the roots of iron deficient cucumber root at pH=5.5



• At pH=5.5, no significant difference can be found in the Fe^{II} formed in 30 min in the case of FeCit, FeEDTA and FeEDDHA. However, the total iron concentration is the highest after applying FeCit.

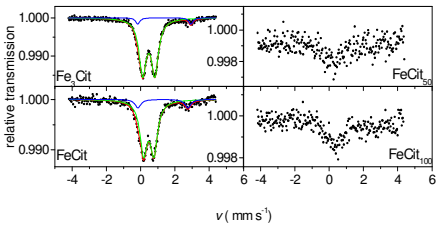
• According to the Mössbauer parameters, no significant difference can be found in the Fe^{III} species accumulated in the root of FeEDTA and FeEDDHA treated plants.

• The Mössbauer parameters of the Fe^{II} component (δ=1.30(2) mms^{-1} Δ=2.93(4) mms^{-1} LW=0.48(7) mms^{-1}) are slightly different from those of Fe^{II}-hexaaqua complex which may indicate the coordination of Fe^{II} to EDTA and EDDHA ligands.

• The significantly lower Fe^{II} concentration at pH=6.5 compared to pH 5.5 can be explained by the lower iron-chelate reductase activity which is in good agreement with literature data [2]. This effect can also explain the low total iron concentration found at high pH values.

• The Mössbauer spectra show that the lower Fe^{II} concentration at higher pH may be due to the change of the Fe^{III}-species accumulated in the root. This suggest a correlation between the iron-chelate reductase activity and the Fe^{III}-complexes formed in the apoplast. However, the formation of new Fe^{III}-species can also be explained by a higher reoxidation rate after reduction at neutral/basic pH.

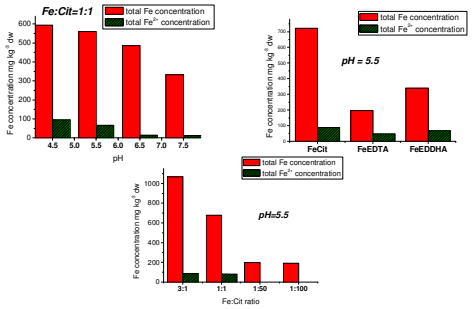
VI. Effect of different Fe:Cit ratios at pH=5.5



• Low Fe:Cit ratio inhibits the accumulation of Fe in the root.

• In the case of Fe_3Cit and FeCit the total Fe^{II} concentration does not change significantly.

VII. Iron concetration of the root*



*The total iron concentration measured by ICP, the total Fe^{2+} concentration was calculated using the Mössbauer data. RSD<5%, n=3

SUMMARY

• In the case of pH above 5.5, a low Fe^{II} concentration was found which resulted also in low total iron concentration of the root.

• In the case of FeCit at pH=7.5, the presence of new Fe^{III} species is favored compared to pH=4.5. This may indicate the formation of hydrous ferric oxides/hydroxides or the structural change of Fe^{III}-carboxylates.

• No significant change was found in the relative amount of Fe^{II} formed and in the Fe^{III} species accumulated in the root after applying FeEDTA or FeEDDHA. In the case of EDTA and EDDHA, a complex formation with Fe^{II} may be possible.

• High Cit:Fe^{III} ratio in the solution inhibited the iron uptake of the root at pH=5.5.

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