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Exudation of organic acids by spinach and the mobilization of Cu, Zn and Cd in soil

W. Römer and H. Keller

Institut für Agrikulturchemie, Universität, Von-Siebold-Str. 6, D-37075 Göttingen, Germany; e-mail: uaac@gwdg.de

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Abstract

The aim of the investigation was to study the organic acid exudation intensity of two spinach cultivars grown under a high and a low P supply and the mobilization of Cu, Zn and Cd by different organic acids in a loamy soil. Ten organic acids were identified. Cultivar Tabu showed a significantly higher rate of exudation of oxalate, citrate and malate than cv. Monnopa. At low P level both cv. showed a significantly higher exudation rate of oxalate and citrate than at high P level. Citric and malic acid were the most efficient organic acids in increasing the solubility of Cu, Zn and Cd. Besides the influence of pH on Zn and Cd solubility, citrate ions further increased the solubility of both elements probably by forming chelates.

Introduction

There are significant differences in the net uptake rates of Cu, Zn and Cd per unit root length (inflow) between spinach cultivars (Keller, 2000) especially under P deficiency. For the cv. Tabu as well as higher inflow rates as compared with cv. Monnopa higher concentrations of Cu, Zn and Cd were also present in the soil solution in which the plants where grown. The reason for this higher heavy metal solubility under cv. Tabu may be a higher exudation rate of organic acids by this cultivar. The first aim of the investigation was therefore to study the exudation intensity of the two cultivars grown under a high and a low P supply and secondly to study the influence of organic acids on the Cu, Zn and Cd solubility compared with the effect of H^+ from HNO_3 on a loamy soil.

Material and methods

Exudation experiment

Spinach plants (cv. Tabu, cv. Monnopa) were grown in quartz sand (2.5 L pots) for 35 days in a growth chamber at two P levels (6 or 12 mg P kg^{-1}). Roots were carefully washed out and transferred into aqua dest. for 1 hour. Afterwards this water was renewed and root exudates were collected within 2 hours. Solutions were prepared according to Gerke (1995) and analysed by HPLC (Waters Corp., Milford, USA). Root length was determined according to Newman (1966). Exudation rate was calculated as nmol cm^{-1} h^{-1}.

Experiment for testing the effect of organic acids on Cu, Zn and Cd solubility in soil

Based on the measured and calculated exudation within 24 hours and a diffusion distance of exudates from roots into soil of 0.75 mm 16 g soil were incubated with 3.2 ml solution of varying concentrations of different organic acids for 6 hours. Afterwards 40 ml distilled water were added and the suspension was shaken for 20 min. The concentrations of the elements were measured in the filtrates using atomic absorption spectroscopy. Soil (Luvisol) characterics: sand 50 g kg^{-1}, silt 750 g kg^{-1}, clay 200 g kg^{-1}, C_{org} 11 g kg^{-1}, pH 7.0 ($CaCl_2$), Cu_{tot} 12, Zn_{tot} 44, Cd_{tot} 0.2 mg kg^{-1}.

Results

Table 1 shows the exudation rates from roots. In both cv. quantitatively the most important anion was oxalate followed by citrate and malate. All the others were present in only low concentrations. Cv. Tabu showed significantly higher rates of exudation of oxalate, citrate and malate than cv. Monnopa. At low P level both cv. showed significantly higher rates of exudation of oxalate and citrate than at high P level.

Figure 1 shows the effect of 4 organic acids with increasing concentrations and decreasing pH on the solubility of Cu, Zn and Cd in soil. Citric and malic acid were the most efficient organic acids in increasing solubility. Whereas the pH between 7 and 5 had no effect on Cu solubility, there was a clear effect of pH in raising Zn and Cd solubility. Besides this influence of pH on Zn and Cd solubility, however, citrate ions further increased the solubility of both elements probably by forming chelates.

Conclusions

The higher inflow of Cu, Zn and Cd by cv. Tabu compared with cv. Monnopa corresponded to its higher exudation of organic acids especially under low P supply and the increased solubility of Cu, Zn and Cd caused by a lower pH and probably by chelation of the 3 elements with citrate and malate anions

Acknowledgement

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References

Gerke J 1995 Habilitationsschrift, Universität Göttingen, Cuvillier Verlag Göttingen

Keller H 2000 PhD-Thesis http://kluedo.ub.uni-kl.de/Chemie/Metadaten/dissertation_33.html

Newman E 1966 J. Appl. Ecol. 3, 133-145.

Table 1. Organic acid exudation of cv. Tabu and Monnopa depending on P nutrition

	Monnopa		Tabu	
	+P	-P	+P	-P
	[nmol cm^{-1} h^{-1}]		[nmol cm^{-1} h^{-1}]	
Oxalic acid	1.08 ^{a}	1.70 ^{b}	2.76 ^{c}	3.91 ^{d}
Citric acid	0.22 ^{a}	0.31 ^{b}	0.69 ^{c}	0.95 ^{d}
2-Oxoglutaric acid	0.01 ^{a}	0.01 ^{a}	0.01 ^{ab}	0.02 ^{b}
Malic acid	0.31 ^{a}	0.36 ^{a}	0.63 ^{b}	0.68 ^{b}
Pyruvic acid	0.03 ^{a}	0.06 ^{ab}	0.06 ^{ab}	0.08 ^{b}
Malonic acid	0.05 ^{a}	0.26 ^{b}	0.08 ^{a}	0.25 ^{b}
Succinic acid	0.12 ^{a}	0.17 ^{ab}	0.22 ^{b}	0.29 ^{b}
Lactic acid	0.27 ^{a}	0.53 ^{b}	0.45 ^{b}	0.48 ^{b}
Acetic acid	0.07 ^{a}	0.03 ^{a}	0.11 ^{b}	0.13 ^{b}
Fumaric acid	0.01 ^{a}	0.02 ^{a}	0.02 ^{a}	0.02 ^{a}

Tartaric, t-aconitic, formic and glutaric acids were not detectable; a, b, c, d show significant differences at $p < 0.05$, Tukey

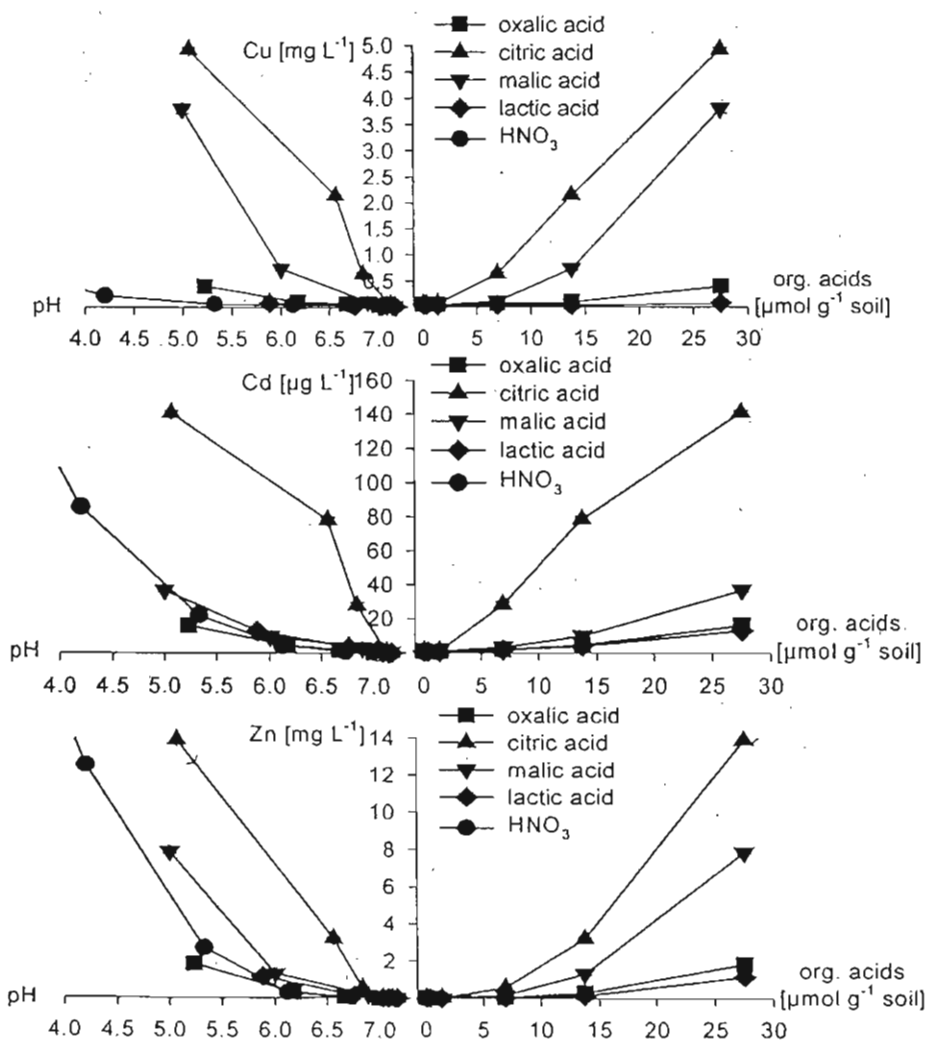

Figure 1. Influence of four organic acids and HNO_3 on Cu, Zn and Cd solubility (standard deviations were smaller than symbols)

Exudation of Organic Acids by Spinach and the Mobilization of Cu, Zn and Cd in Soil

W. Römer, H. Keller
Institut für Agrikulturchemie, University of Goettingen, Von Siebold–Str. 6, D-37075 Goettingen, Germany, uaaci@gwdg.de

ABSTRACT *The aim of the investigation was to study the organic acid exudation intensity of two spinach cultivars grown under high and low P supply and to measure the mobilization of Cu, Zn and Cd by 4 different organic acids and HNO_3 in a neutral loamy soil. Cultivar Tabu showed a significantly higher rate of exudation of oxalate and citrate than cv. Monnopa. At low P level both cv. showed a significantly higher exudation rate of oxalate and citrate than at high P level. Amoung the 4 organic acids, citric and malic acid were the most efficient organic acids in increasing the solubility of Cu, Zn and Cd.*

INTRODUCTION

Under P deficiency, 2 spinach cultivars showed significant different Cu, Zn and Cd net uptake rates (Keller, 2000). The reason for this result may be an excretion of organic acids which increase the solubility of these heavy metals in the rhizosphere of the plants. Therefore we investigated:
1- the organic acid exudation of the two cultivars at 2 P levels and,
2- the effect of four organic acids and HNO_3 on the Cu, Zn and Cd solubility in a Luvisol.

EXUDATION EXPERIMENT

Spinach cultivars Tabu and Monnopa were grown in quartz sand for 35 days at two P levels (6 or 12 mg P kg^{-1}). Roots were carefully washed out and transferred into aqua dest. for collecting root exudates analyzed by HPLC. Root length was determined and exudation rate per unit root length was calculated.

RESULTS

- Oxalic, citric, malic and lactic acid were quantitatively the most excreted organic acids by both cultivars. (Table 1).
- Cultivar Tabu showed significantly higher exudation rates of oxalic, citric and malic acid than Monnopa.
- At low P level, both cultivars increased significantly the exudation rate of oxalic and citric acid.

Table 1 : Organic acid exudation of cv. Tabu and Monnopa depending on P supply

	Monnopa		Tabu	
	+ P	- P	+ P	- P
	nmol cm^{-1} h^{-1}			
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Tartaric, fumaric, formic and glutaric acids were not detectable. a, b, c, d show significant differences at p < 0.05 Tukey.

CONCLUSION

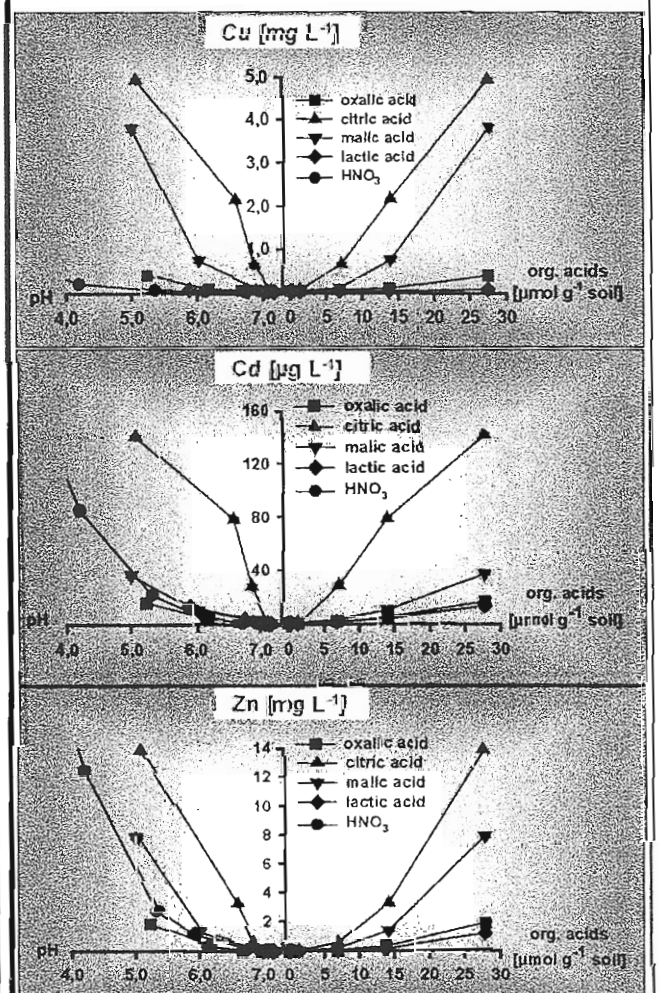
The higher inflow of Cu, Zn and Cd by cv. Tabu compared with cv. Monnopa corresponded to its higher exudation of organic acids especially under low P supply and the increased solubility of Cu, Zn and Cd caused by a lower pH and probably by chelation of the 3 elements with citrate and malate anions.

EXPERIMENT FOR TESTING THE EFFECT OF ORGANIC ACIDS ON Cu, Zn, AND Cd SOLUBILITY IN SOIL

16 g Luvisol were incubated with 3.2 ml solution of varying concentrations of different organic acids or HNO_3 for 6 h. Afterwards 40 ml distilled water were added and the suspension was shaken for 20 min. Cu, Zn and Cd concentrations were measured in the filtrates by using AAS. Soil characteristics are: sand 5 %, silt 75 %, clay 20 %, C_{org} 1.1 %, pH $_{(CaCl_2)}$ 7.0, Cu_{tot} 12, Zn_{tot} 44, Cd_{tot} 0.2 mg kg^{-1}.

RESULTS

- Citric and malic acid were the most efficient anions by increasing Cu, Zn and Cd solubility in the soil.
- Whereas the pH between 7 and 5 had no effect on Cu solubility, there was a clear effect of pH increasing Zn and Cd solubility.
- Besides the influence of pH on Zn and Cd solubility, citrate ions further increased the solubility of both elements probably by forming chelates.



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