Synthesis and Use of Iron Humates for Correction of Iron Deficiency Chlorosis in Higher Plants

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1. INTRODUCTION

Deficiency of bioavailable iron in soils leads to plant disease known as chlorosis. The most efficient way to correct iron chlorosis is to use synthetic iron (III) chelates such as DTPA and EDDHA. However, application of these chelates may bring about negative consequences for environment as their migration along the soil profile. Therefore, development of iron chelates based on a use of natural macroligands is of great importance. From this prospective, humic substances (HS), which contain large amounts of carboxyl and hydroxyl groups seem to be the most promising compounds. The objective of this study was to develop humic-based iron chelates and to evaluate their efficiency for use as chlorosis correctors.

2. MATERIALS AND METHODS

Iron Humates: Three samples of iron humates were prepared from commercially available potassium humate derived from leonardite (Sakhalin Humate^{TM}) and iron(II) sulfate at different pH in the presence and absence of ascorbic acid. The insoluble part of potassium humate was separated with centrifugation. Iron(II) sulfate with concentration 725 mmol/l was added to 5% solution of potassium humate at pH 11 and 9. The solution of ascorbic acid (2%) was used for stabilization of Fe^{2+} oxidation state. The obtained solution was dried on rotary evaporator. Iron content was determined spectrophotometricaly using complexation with o-phenantroline after oxidative wet digestion. For determination of redox state of iron species, the obtained samples were investigated using Mössbauer spectrometry.

Bioassay: Wheat plants (*Triticum aestivum* L. var. Krestianka) were used as biotarget and Knopp nutrient solution without iron was used as a control. Wheat seeds were germinated in distilled water in the dark at 25°C for 72 hours. Seedlings were grown on

Knopp nutrient solution with iron supplied in the different forms at the concentration of 25 μmol/l. The commercially available chelate Sequestren Fe-EDDHA was used as a positive control. Length of shoots and roots, mass of roots, iron and chlorophyll content in the shoots were measured. Chlorophyll was extracted from shoots with acetone. The contents of a and b chlorophyll were determined spectrophotometricaly. Iron content was determined after oxidative digestion of dried shoots using o-phenantroline technique.

3. RESULTS AND DISCUSSION

The content of iron in all three preparations was 8.8±0.3%, solubility of samples synthesized at pH 9 in the presence of ascorbic acid (AA) was 127±4 g/l, in the absence of AA it was 52±4 g/l. The sample obtained at pH 11 turned out insoluble. Mössbauer spectra of samples synthesized at pH 9 have shown that in the absence of AA, all iron present in iron humate was Fe(III), whereas in the sample obtained in the presence of ascorbic acid, Fe(III) accounted for 94% and Fe(II) - 6%.

The results of bioassay have shown that the both samples of iron humates obtained at pH 9 caused stimulating effect on wheat seeds: the root weight for sample with AA was 112 ± 4 % of control and for sample without AA it was 109±4% of control. At the same time, for Fe-EDDHA the value of this parameter was 106±4% of control. Both Fe-EDDHA and all soluble iron humates promoted accumulation of chlorophyll in shoots. Another indicator of the efficiency of the sample as chlorosis corrector is iron content in shoots. The highest iron content was obtained for iron humate synthesized at pH 9 with AA (243±9 mg/kg that corresponds to 430% of control) and lesser content - for sample without AA (170±7 mg/kg that corresponds to 250% of blank). Of importance is that both iron humates outcompeted Fe-EDDHA: the iron content accounted for 117±6 that corresponds to 173% of blank.

4. CONCLUSIONS

The soluble iron humates with content of iron >8% were prepared. Application of ascorbic acid increased solubility of iron humate and facilitated iron accumulation by plants. The obtained iron humates were shown to be efficient chlorosis correctors.

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