The Reduction of Fe(III) to Fe(II) and V(V) to V(IV) by Polygalacturonic Acid: a Reduction and Complexation Mechanism of Biochemical Significance

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Sugar–cation associations could have important biological implications because metal complexation is likely to occur on the surfaces of polysaccharides, especially on cell walls [1].

An extracellular apparatus rich in acid polysaccharides is essential for plants, as it allows the roots to remove micronutrients from clay particles through contact cation exchange [2].

As part of our research effort toward understanding the interaction of metal ions with polyuronic acids, we have recently reported data for some metal complexes formed by polygalacturonic acid [3, 4]. The study has shown that the metal binding is governed by selective mechanisms. Indeed, interactions of an electrostatic nature allow some ions to retain the inner hydration shell, whereas inner-sphere binding through carboxylate groups produces immobilization of ions such as Cu^{2+} and VO^{2+}.

To obtain information about the mechanisms governing the iron retention, we prepared iron(II) and (III) polygalacturonates and characterized them by means of Mössbauer and ESR spectroscopies [5]. Interestingly, Mössbauer spectra of the Fe^{3+}-exchanged samples revealed the presence of both Fe(II) and Fe(III) ions bound to the polymeric matrix [5].

As plant roots reduce Fe(III) to Fe(II) to make available this element [6], we have paid attention to reduction processes occurring upon interaction of polygalacturonic acid with some mineral species. The understanding of the pertinent mechanisms could contribute to our knowledge of processes of biochemical significance, particularly in relation to the mineral nutrition and the survival of plants.

Results and Discussion

Both Fe(II) and Fe(III) ions are shown by the Mössbauer spectra of samples obtained upon interaction of polygalacturonic acid with Fe^{3+} solutions (Fig. 1). Unlike the Fe(III)-parameters, which do not vary significantly with differing hydration degrees of the samples or pH values of the suspensions, the Fe(II) parameters are dependent on the water content of the samples, as previously observed for iron polygalacturonates [5].

After suspending the polyacid in aqueous solution of $NaVO_3$, V(IV) ions are formed, as shown by ESR spectroscopy (Fig. 2). The reduction is rather fast, but a limiting value is attained by the signal amplitude even in the presence of excess of VO_3^-. The ESR hyperfine pattern indicates a single oxovanadium(IV) species rigidly bound to the polysaccharide matrix through carboxylate groups [3], and excludes the presence of V(IV) in solution.

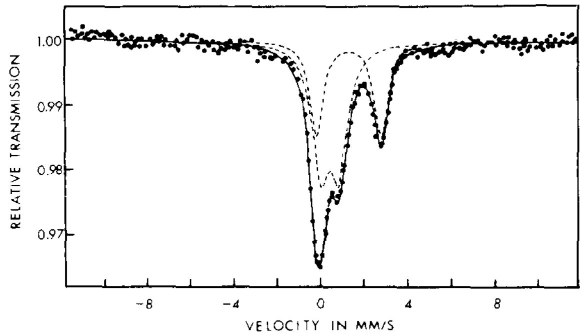

Fig. 1. Mössbauer spectrum of Fe(III)-exchanged polygalacturonate ($H_2O \sim 15$ wt.%, Fe ~ 2.4 wt.%).

Fig. 2. Room temperature X-band e.s.r. spectrum of the suspension of polygalacturonic acid in $NaVO_3$ solution.

To give explanation of such processes, analytical tools have been employed. The reaction involving VO_3^- was easily investigated because of the absence of V(IV) in solution. Thus, the amount of reduced vanadium was estimated as difference between the amount of V initially added and that remaining in solution after removal of the solid phase (see, *e.g.*, Table I). The quantitative analysis showed that almost four vanadium atoms were reduced per reducing end-unit of the polysaccharide. Finally,

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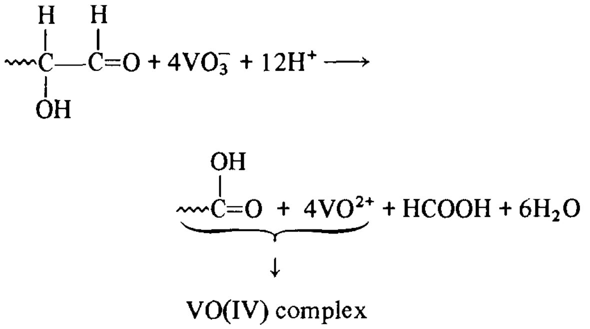
TABLE I. Representative Data Concerning the Interaction of PGA (1.0 g) with $NaVO_3$ Solutions.

VO_3^- added (mol)	VO_3^- remaining in solution (mol)	V reduced (mol)	HCOOH formed (mol)
9.44×10^{-4}	1.60×10^{-4}	7.84×10^{-4}	1.80×10^{-4}
18.94×10^{-4}	11.06×10^{-4}	7.88×10^{-4}	1.75×10^{-4}

formic acid was detected and determined in solution by ion exchange chromatographic analysis (Fig. 3). In any case a stoichiometric amount, almost one mol of formic acid per reducing end-unit of the polysaccharide, seemed to be formed.

Based on these results it is suggested that the reaction mechanism involves oxidation of the terminal units of the polymeric chains through a four-electron process:



$$\sim\!\!\sim\overset{H}{\underset{OH}{C}}-\overset{H}{C}=O + 4VO_3^- + 12H^+ \longrightarrow \underbrace{\sim\!\!\sim\overset{OH}{C}=O + 4VO^{2+}}_{\downarrow \atop \text{VO(IV) complex}} + HCOOH + 6H_2O$$

Further confirmation of this mechanism is given by the absence of reducing properties in the the polyuronate after the reaction.

Preliminary results indicate that the reduction of iron(III) also takes place through the formation of formic acid, and the analytical results will be reported in a further paper.

The reduction of Fe(III) at the plant roots is believed to be due to the release of specific reductants, several inferences having been drawn [6]. Our results indicate that polyuronates, which are main components of the roots and are responsible for root–soil interactions, are also active in these redox processes. These mechanisms are likely required in order to transform rigidly bound tervalent species into faster-moving divalent ions.

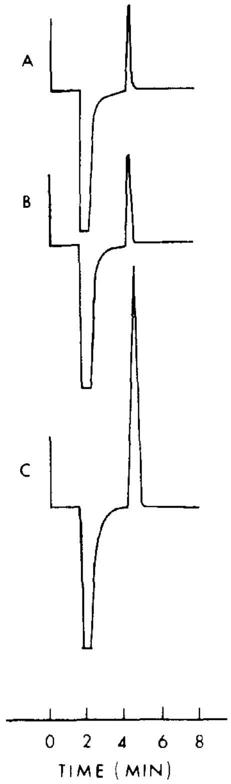


Fig. 3. Detection of formic acid by chromatographic analysis: A: standard solution of formic acid; B: aqueous phase after reduction of VO_3^- by polygalacturonic acid; C: sample B enriched with formic acid.

Experimental

Polygalacturonic acid (PGA), obtained from Baker, was purified according to described methods [7]. Reducing terminal units ($\sim 2.04 \times 10^{-4}$ mol/g PGA) were determined according to the method described by Becher and Englis [8].

Reduction experiments were performed by contacting PGA (~0.5 g) with $NaVO_3$ (or $Fe(NO_3)_3$) aqueous solutions (pH *ca.* 4.5 and 3.0, respectively). The final volume was adjusted to *ca.* 20 cm^3. The suspensions were allowed to stand overnight at 5–10 °C and then dialyzed against deionised water in order to remove all the soluble products. Vanadium was determined in solution using a Spectraspan IV emission spectrometer. Formic acid was determined by ion exchange liquid chromatography using a Wescan 260 ion analyser equipped with a Wescan 269-001 anion column. A 4 m*M* potassium hydrogen phthalate (pH 4.5) solution was used as eluant. The flow rate was 2 cm^3 min^{-1}.

ESR and Mössbauer spectra were obtained as previously described [3, 5].

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References

1 S. J. Angyal, *Chem. Soc. Rev.*, *10*, 415 (1981).
2 S. Ramamoorthy and G. G. Leppard, *J. Theor. Biol.*, *66*, 527 (1977).
3 S. Deiana, L. Erre, G. Micera, P. Piu and C. Gessa, *Inorg. Chim. Acta*, *46*, 249 (1980).
4 S. Deiana, G. Micera, G. Muggiolu, C. Gessa and A. Pusino, *Colloid Surface*, *6*, 17 (1983).
5 G. Micera, S. Deiana, C. Gessa and M. Petrera, *Inorg. Chim. Acta*, *56*, 109 (1981).
6 J. C. Brown, 'Genetically Controlled Chemical Factors Involved in Absorption and Transport of Iron by Plants', in 'Bioinorganic Chemistry', K. Raymond Ed., Vol. 2., *Adv. Chem. Ser.*, *162*, Amer. Chem. Soc., Washington D.C., pp. 93–103 (1977).
7 H. H. G. Jellinek and P.-Y. A. Chen, *J. Polym. Sci.*, *A1 10*, 287 (1972).
8 H. C. Becker and D. T. English, *Ind. Eng. Chem., Anal. Ed.*, *13*, 15 (1941).