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INTERACTION BETWEEN POLYHYDROXY COMPOUNDS AND VANADATE IONS: ELECTROPHORESIS AND COMPOSITION OF COMPLEXES

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ABSTRACT

Several polyhydroxy compounds form anionic complexes with vanadate ions. The V/polyol ratio in several complexes has been determined. In the pH range 5–9.5, the complexing agent is likely to be a metavanadate ion, $(VO)_n^{n-}$. Electrophoresis in sodium metavanadate solution is particularly useful for the separation of hexitols, hex-2-uloses, and several disaccharides.

INTRODUCTION

The anions derived from the transition metals of group VA and VIA condense reversibly as the pH of their solution is lowered, to give a series of anions of isopoly-acids[1]. The dimolybdate ($Mo_2O_7^{2-}$) and ditungstate ions ($W_2O_7^{2-}$), although not normally present in molybdate and tungstate solutions, respectively, form complexes with stereochemically defined polyhydroxy compounds which form the basis of separatory methods[2-4]. Whereas the condensations of the molybdate (MoO_4^{2-}) and tungstate ions (WO_4^{2-}) occur at pH 7 and lower[1], the condensation of the orthovanadate ion (VO_4^{2-}) commences at pH 11. Frahn and Mills[5] briefly reported that vanadate ions complex with D-glucitol and D-mannitol. We now report on the properties of the complexes that are formed from vanadate ions and polyhydroxy compounds.

RESULTS AND DISCUSSION

D-Glucitol showed maximal electrophoretic mobility in vanadate solution at pH ~8, whereas maximal changes in the optical rotatory power occurred at pH 5.9 (Fig. 1). The optical rotatory power of solutions containing D-mannitol or maltose and sodium metavanadate were similarly affected. As metavanadate exists in the pH range 5.5–9.5, it is unlikely that the two phenomena involve two different types

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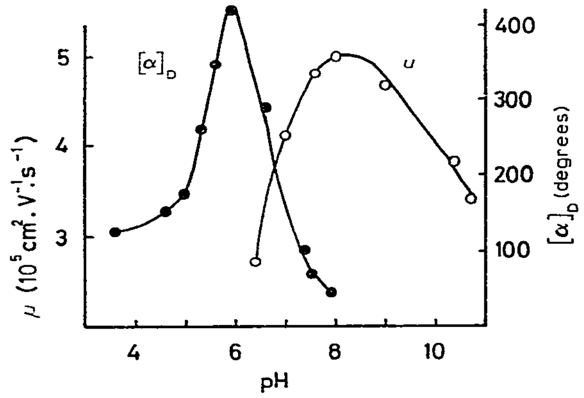

Fig. 1. Effect of pH on electrophoretic mobility (*u*) and $[\alpha]_D$ of D-glucitol in vanadate solutions.

of vanadate ions, but they may be due to different degrees of ionisation of the complex. Although pH ~8 may not be optimal for electrophoresis of all compounds, the electrophoretic mobilities, expressed relative to that of D-glucitol, have been determined by using 1.5% aqueous sodium metavanadate (pH ~8.7).

In contrast to the complexes formed with other anionic reagents[3] or cations[6,7], it is apparent that complex formation with vanadate cannot be attributed to a single, stereochemically defined polyol grouping. For example, replacement of a hydroxyl group in alditols and aldoses by a hydrogen atom or a methoxyl group may lower the electrophoretic mobility (*cf.* D-glucitol and 3-deoxy-L-*xylo*-hexitol) or not affect it greatly (*cf.* D-glucose and 2-*O*-methyl-D-glucose), but replacement of the hydrogen atom in that hydroxyl group by a D-glucopyranosyl group may enhance the electrophoretic mobility (*cf.* D-glucitol and 3-*O*-α-D-glucopyranosyl-L-gulitol, and D-glucose and kojibiose). It therefore seems that both moieties of the glucopyranosylhexitols and disaccharides provide hydroxyl groups for complex formation and that the complexing vanadate ion bridges the glucosidic oxygen atom.

Affinity for vanadate therefore cannot be regarded as diagnostic for the stereochemistry of a particular polyol grouping (*cf.* affinity[3,7] for $Mo_2O_7^{2-}$, $W_2O_7^{2-}$, and Ca^{2+}). Nevertheless, several compounds that are difficult to separate by other methods have sufficiently different $M_S(V)$ values to allow their separation and distinction. Pertinent examples are the six hexitols, hex-2-uloses, α-D-glucopyranosyl-D-fructoses, and, particularly, the α and β isomers of the D-glucopyranosyl-D-glucoses (*cf.* kojibiose and sophorose, nigerose and laminaribiose, and maltose and cellobiose). Electrophoresis in sodium metavanadate solution is therefore complementary to other separatory methods.

The ratios of V/polyol in the complexes with D-glucitol, D-mannitol, galactitol, 3-*O*-α-D-glucopyranosyl-L-gulitol, D-ribose, and maltose were determined by Job's method of continuous variation[8,9], *i.e.*, in these cases, by measurement of either optical rotations (α) and/or absorbances (*A*) of a series of reaction mixtures in which the sum of the molar proportions of sodium metavanadate and polyhydroxy compound is kept constant. Measurements on solutions containing vanadate were made after adjustment to pH 5.9, at which value the reaction mixtures exhibited maximal changes in optical rotatory power. At each concentration, the optical rotatory power of the polyol-blank or the absorbance of the vanadate-blank were subtracted from those of the reaction mixtures. The resulting values, $\Delta\alpha$ and ΔA, represent the increase in optical rotatory power and absorbance, respectively, produced by the vanadate–polyol complex, and maximum $\Delta\alpha$ and ΔA occur when the amounts of

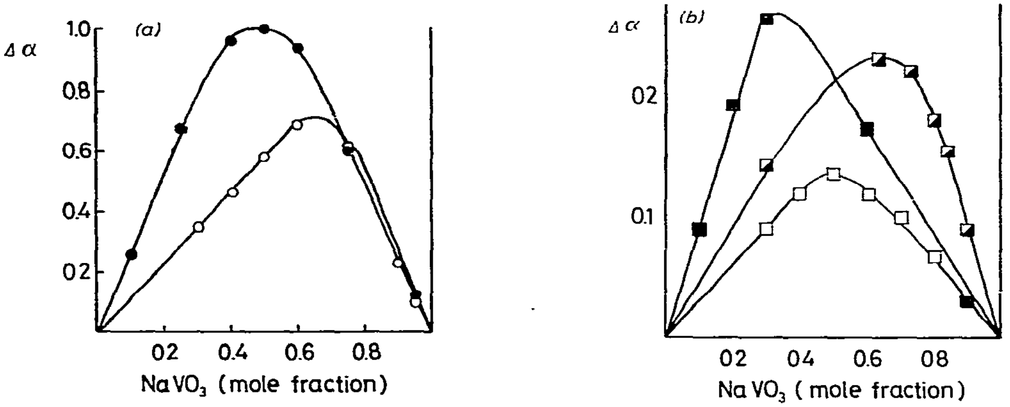

Fig. 2. Plots of $\Delta\alpha$ *versus* relative concentrations of vanadate and polyhydroxy compounds: (*a*) ○, D-glucitol (0.2M); ●, D-mannitol (0.5M); (*b*) □, 3-*O*-α-D-glucopyranosyl-L-gulitol (0.5M); ■, D-ribose (0.3M); ◪, maltose (0.1M).

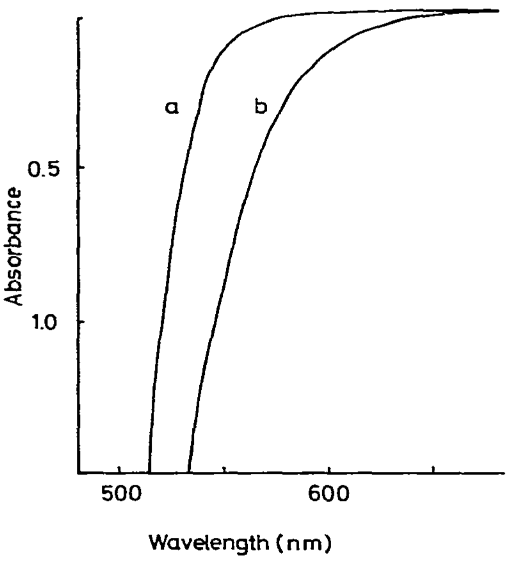

Fig. 3. Absorption edges of (*a*) a solution of $NaVO_3$ adjusted to pH 5.9, and (*b*) a solution containing $NaVO_3$ and D-glucitol adjusted to pH 5.9.

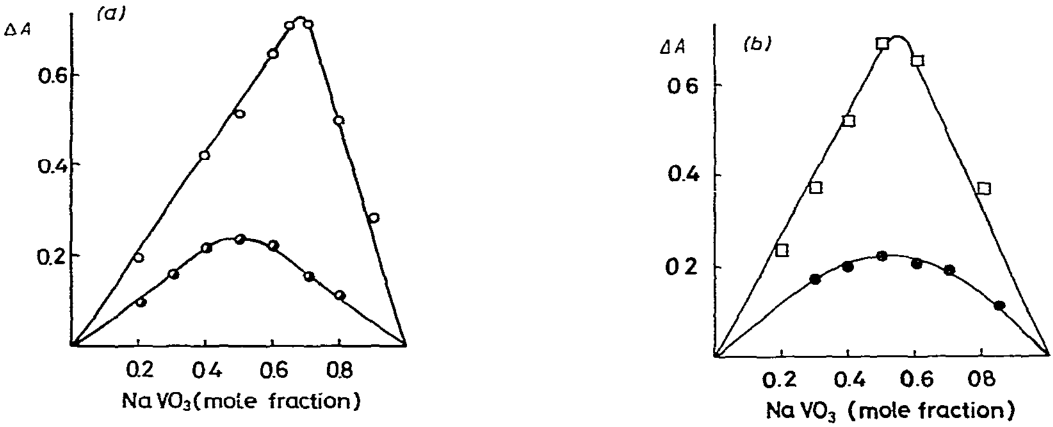

Fig. 4. Plots of ΔA *versus* relative concentrations of vanadate and polyhydroxy compounds: (*a*) ○, D-glucitol (0.2M, 550 nm); ◐, galactitol (0.2M, 510 nm); (*b*) ●, D-mannitol (0.2M, 460 nm); □, 3-*O*-α-D-glucopyranosyl-L-gulitol (0.025M, 410 nm).

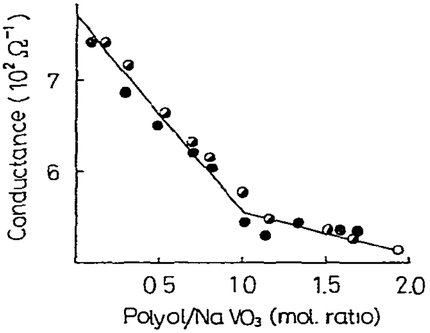

Fig. 5. Conductimetric titration of sodium metavanadate: ●, with D-mannitol; ◐, with galactitol.

vanadium and polyol used are in the same ratios as those in which they appear in the complex.

The wavelengths for measurements of absorbances were selected from the shifts of the absorption edges of vanadate solutions, occasioned by the addition of the polyhydroxy compounds. The results obtained with D-glucitol are shown in Fig. 3. The wavelengths selected from such measurements with the six compounds studied are indicated in the legend to Fig. 4.

The results obtained by the method of continuous variation (Figs. 2 and 4) show that the V/polyol ratios in the complexes with galactitol, D-glucitol, D-mannitol, 3-*O*-α-D-glucopyranosyl-L-gulitol, D-ribose, and maltose are 1, 2, 1, 1, 0.5, and 2, respectively. Conductimetric titration of sodium vanadate with galactitol or D-mannitol (Fig. 5) also showed that the V/polyol ratios in the complexes with these hexitols are 1. The fact that, in several cases (galactitol, D-glucitol, D-mannitol, and 3-*O*-α-D-glucopyranosyl-L-gulitol), measurements of different physical properties (*i.e.*, optical rotation, absorbance, and conductivity) gave identical results shows

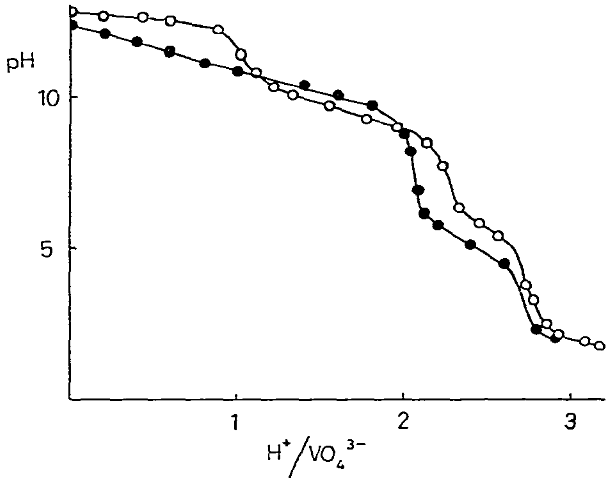

Fig. 6. Potentiometric titrations of orthovanadate solutions with sulphuric acid: ○, sodium orthovanadate; ●, D-glucitol and sodium orthovanadate in the molar ratio 7.5:1.

that the changes in these properties were effected by the same complex. These observations strengthen the above conclusions.

The steps involved in the formation of the isopolyvanadates have been deduced from physicochemical studies[10], *e.g.*, potentiometric titration of sodium orthovanadate solution with sulphuric acid. Inflexion points in the titration curve occurred at H^+/VO_4^{3-} ratios of 1, 2, and 2.6, and these have been attributed to the formation of $V_2O_7^{4-}$, $V_4O_2^{4-}$, and $H_2V_{10}O_{28}^{4-}$, respectively (Eq. *1–3*). These results are essentially reproducible (Fig. 6).

$$2\,H^+ + 2\,VO_4^{3-} \rightarrow V_2O_7^{4-} + H_2O \qquad (1)$$

$$2\,H^+ + 2\,V_2O_7^{4-} \rightarrow V_4O_{12}^{4-} + H_2O \qquad (2)$$

$$12\,H^+ + 5\,V_4O_{12}^{4-} \rightarrow 2\,H_2V_{10}O_{28}^{4-} + 4\,H_2O \qquad (3)$$

However, when the titration was carried out in the presence of excess of D-glucitol (*i.e.*, V/polyol $\leqslant 2$), no inflexion occurred at a H^+/VO_4^{3-} ratio of 1. It is apparent that, in the pH range 12.5–9.5, the orthovanadate ion forms a complex with D-glucitol, and that, as a result, the divanadate ion is not formed (*cf.* Eq. *1*) in detectable amounts. In the pH range 9.5–5, the complexing ion is produced from a H^-/VO_4^{3-} ratio of 2. It is therefore likely that it is a metavanadate ion, which may be formulated as VO_3^-, $V_2O_6^{2-}$, $V_3O_9^{3-}$, $V_4O_{12}^{4-}$, *i.e.*, $(VO_3)_n^{n-}$ formed according to Eq. *4*.

$$2n\,H^+ + n\,VO_4^{3-} \rightarrow (VO_3)_n^{n-} + n\,H_2O \qquad (4)$$

This uncertainty prevents unambiguous assignments of structures to the vanadate–polyol complexes. However, the non-specific complexing with vanadate (*cf.* electrophoresis) and the various ratios of V/polyol in the complexes suggest that the value of *n* of the complexing metavanadate ion may depend on the polyol ligand.

TABLE I

ELECTROPHORETIC MOBILITIES OF POLYHYDROXY COMPOUNDS IN SODIUM METAVANADATE SOLUTION

Compound	Ms(V)[a]
Compounds having Ms (V) $\geqslant$ *0.1*	
Erythritol	0.1–0.2
D-Threitol	0.1–0.2
D-Arabinitol	0.32
1-deoxy-	0.26
D-Lyxitol	0.36
1-deoxy-	0.11
Ribitol	0.31
Xylitol	0.60
1-deoxy-	0.48
Allitol	0.70
D-Altritol	0.98
1-deoxy-	0.67
1,6-dideoxy-	0.34
2-Deoxy-D-*arabino*-hexitol	0.12
3-Deoxy-D-*arabino*-hexitol	0.11
2-Deoxy-D-*lyxo*-hexitol	0.54
3-Deoxy-D-*ribo*-hexitol	0.11
2-Deoxy-L-*xylo*-hexitol	1.15
3-Deoxy-L-*xylo*-hexitol	0.33
Galactitol	0.48
1-deoxy-L-	0.33
1,6-dideoxy-	0.34
D-Glucitol	1.00
3-*O*-β-D-glucopyranosyl-	0.33
L-Gulitol; 1-*O*-α-D-glucopyranosyl-	0.76
1-*O*-β-D-glucopyranosyl-	0.74
3-*O*-α-D-glucopyranosyl-	1.52
3-*O*-β-D-glucopyranosyl-	0.77
1-*O*-methyl-	0.68
L-Iditol	1.50
D-Mannitol	0.40
1,6-dideoxy-	0.20
2,5-*O*-methylene-	0.14
1,2-di-*O*-methyl-	0.34
2,5-di-*O*-methyl-	0.11
D-Talitol; 1-deoxy-	0.37
D-Lyxose	0.10
D-Ribose	0.40
D-Xylose	0.10
2-Deoxy-D-*arabino*-hexose	0.11
2-Deoxy-D-*lyxo*-hexose	0.16
5-Deoxy-D-*xylo*-hexose	0.79
D-Glucose	0.10
6-deoxy-	0.10
2-*O*-methyl-	0.10
2-*O*-α-D-glucopyranosyl- (kojibiose)	2.9
2-*O*-β-D-glucopyranosyl- (sophorose)	0.13

TABLE I (continued)

Compound	Ms(V)[a]
3-*O*-α-D-glucopyranosyl- (nigerose)	2.9
3-*O*-β-D-glucopyranosyl- (laminaribiose)	0.10
4-*O*-α-D-glucopyranosyl- (maltose)	0.26
6-*O*-α-D-glucopyranosyl- (isomaltose)	0.10
6-*O*-β-D-glucopyranosyl- (gentiobiose)	0.10
D-Gulose	0.17
3-Deoxy-D-*erythro*-hex-2-ulose	0.10
D-*arabino*-Hex-2-ulose (D-fructose)	0.45
1-*O*-methyl-	0.19
3-*O*-α-D-glucopyranosyl- (turanose)	0.26
4-*O*-α-D-glucopyranosyl- (maltulose)	0.31
5-*O*-α-D-glucopyranosyl- (leucrose)	0.45
6-*O*-α-D-glucopyranosyl- (isomaltulose)	0.35
D-*lyxo*-Hex-2-ulose (D-tagatose)	0.39
D-*xylo*-Hex-2-ulose (D-sorbose)	0.2–0.8
Compounds having Ms(V) < *0.1*	
Glycerol	
2-Deoxy-D-*erythro*-pentitol	
D-Glucitol; 2-*O*-α-D-glucopyranosyl-	
2-*O*-β-D-glucopyranosyl-	
3-*O*-α-D-glucopyranosyl-	
L-Arabinose	
methyl β-pyranoside	
2-Deoxy-D-*erythro*-pentose	
methyl β-pyranoside	
D-Galactose	
L-Galactose; 6-deoxy-	
D-Glucose; 4-*O*-β-D-glucopyranosyl- (cellobiose)	
3-*O*-methyl-	
6-*O*-methyl-	
D-Mannose	

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[a] Electrophoretic mobility is expressed relative to that of D-glucitol.

EXPERIMENTAL

Electrophoresis. — The electrolyte was 1.5% aqueous sodium metavanadate (pH ∼8.7). Electrophoretic mobilities were determined at ∼45 V.cm^{-1} with Whatman No. 3MM paper, 5-hydroxymethyl-2-furaldehyde for correction of electro-osmosis, and a saturated solution of potassium permanganate in acetone as staining reagent. Electrophoretic mobility, $M_S(V)$, was expressed relative to that of D-glucitol. The results are shown in Table I.

In one series of experiments, the pH of the electrolyte was adjusted to values in the range 6–11, and the electrophoretic mobilities of D-glucitol were determined. The results are shown in Fig. 1.

Effect of pH on optical rotation of D-*glucitol in vanadate solution.* — Solutions containing D-glucitol (0.2%) and sodium metavanadate (1.25%) were adjusted to pH values in the range 3.8–8 by addition of conc. H_2SO_4. The optical rotations (Fig. 1) are expressed on the basis of the concentration of D-glucitol.

Determination of V/polyol ratio in complex by polarimetry. — Solutions (20 cm^3) containing the polyol and sodium metavanadate in various concentrations (but with the combined concentrations shown in the legend to Fig. 2) were adjusted to pH 5.9 by addition of conc. H_2SO_4 and made up to 25 cm^3. The differences in optical rotatory power [$\Delta\alpha$, using a 1-dm tube, except for D-glucitol (0.25-dm tube)] between these solutions and those containing only the polyhydroxy compound were plotted *versus* the mole fraction of $NaVO_3$ (Fig. 2).

Determination of V/polyol ratio in complex by absorption measurements. — The u.v. and visible spectra of the following mixtures, adjusted to pH 5.9 and then made up to 25 cm^3, were recorded: (*a*) 0.2M D-glucitol (8 cm^3) and 0.2M $NaVO_3$ (12 cm^3); (*b*) 0.2M D-mannitol (10 cm^3) and 0.2M $NaVO_3$ (10 cm^3); (*c*) 0.2M galactitol (5 cm^3) and 0.2M $NaVO_3$ (5 cm^3); (*d*) 0.05M 3-*O*-α-D-glucopyranosyl-L-gulitol (15 cm^3) and 0.05M $NaVO_3$ (5 cm^3), as well as solutions containing only sodium metavanadate of the respective concentrations. The results obtained with D-glucitol are shown in Fig. 3.

Solutions (20 cm^3) containing the polyol and sodium metavanadate in various concentrations (but with the combined concentrations shown in the legend to Fig. 4) were adjusted to pH 5.9 and made up to 25 cm^3. The differences in absorbance (ΔA), at wavelengths shown in Fig. 4, between these solutions and those made up from only sodium metavanadate were plotted against the mole fraction of $NaVO_3$ (Fig. 4).

Conductimetric titration of sodium metavanadate with hexitols. — Weighed quantities of solid D-mannitol (total, 3.474 g) and galactitol (total, 3.488 g) were added to separate solutions of 0.1M $NaVO_3$ (100 cm^3) and the conductivity was measured after each addition. The results are shown in Fig. 5.

Potentiometric titrations of sodium orthovanadate. — Separate solutions containing (*a*) $Na_3VO_4 \cdot 14\,H_2O$ (10.89%) or (*b*) $Na_3VO_4 \cdot 14\,H_2O$ (10.89%) and D-glucitol (2.25%) were titrated with 0.05M sulphuric acid. The results are shown in Fig. 6.

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