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Redox and ligand-exchange chemistry of chromium(VI/V)-methyl glycoside systems †

Marcela Rizzotto,[a] Aviva Levina,[b] Mabel Santoro,[a] Silvia García,[a] María Inés Frascaroli,[a] Sandra Signorella,*[a] Luis F. Sala *[a] and Peter A. Lay *[b]

[a] *Departamento de Química, Facultad de Ciencias Bioquímicas y Farmacéuticas, UNR, Suipacha 531, S2002LRK Rosario, Argentina*
[b] *Centre of Heavy Metals Research, School of Chemistry, University of Sydney, NSW 2006, Australia*

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In order to establish a general pattern for the redox and coordination chemistry of glycosides with Cr(VI) and Cr(V), reactions of a series of methyl glycosides with Cr(VI) and Cr(V) have been studied at different acidities. Oxidations of methyl α- and β-D-glucopyranoside (Glc1Me), methyl α- and β-D-mannopyranoside (Man1Me), methyl α- and β-D-galactopyranoside (Gal1Me) and methyl α- and β-D-ribofuranoside (Rib1Me) by Cr(VI) proceed rapidly at pH ≤ 1, and yield Cr(III) and methyl glycofuranuronolactone as final products when an excess of methyl glycoside over Cr(VI) is used. At constant $[H^+]$, the reaction follows the rate law $-d[Cr(VI)]/dt = k_H$ [Gly1Me] [Cr(VI)]. Relative reactivities of methyl glycosides toward Cr(VI) reduction are: β-Rib1Me > α-Gal1Me > α-Rib1Me ≈ β-Gal1Me > β-Man1Me > α-Man1Me > α-Glc1Me > β-Glc1Me. This sequence is interpreted in terms of the degree of unfavorable steric interactions induced by the nonbonded 1,3-diaxial interactions in the respective Gly1Me-Cr(VI) monochelate, which is formed in rapid equilibrium that precedes the rate determining step. For all the glycosides, the oxidation rate decreases with an increase in pH value and becomes negligible at pH > 5. At pH 5.5 and 7.5, addition of an excess of α-Man1Me or α(β)-Gal1Me to an equimolar cysteine-Cr(VI) mixture, afforded two EPR triplets at g_{iso1} 1.9802 and g_{iso2} 1.9800/1 with A_{iso} 16.5(3) × 10^{-4} cm^{-1} in a 50 : 50 g_{iso1} : g_{iso2} ratio. The EPR spectral parameters and the superhyperfine pattern of the signal are consistent with the presence of two geometric isomers of the $[Cr^{V}O(cis\text{-}O^3,O^4\text{-Gal1Me})_2]^-$ and $[Cr^{V}O(cis\text{-}O^2,O^3\text{-Man1Me})_2]^-$ complexes. The same final spectral pattern is observed at pH 7.5 for the ligand-exchange reaction of Man1Me and Gal1Me with $[Cr^{V}O(ehba)_2]^-$ (ehba = 2-ethyl-2-hydroxybutanoato(2−)). No EPR signal is observed when an excess of Xil1Me or Glc1Me is added to an equimolar cysteine-Cr(VI) mixture. In the ligand-exchange reactions of $[Cr^{V}O(ehba)_2]^-$ at pH 7.5 with Xil1Me or Glc1Me, a very low intensity EPR singlet is observed at g_{iso} 1.9799. These results show that only glycosides with one *cis*-diolato group (such as Man1Me and Gal1Me) are effective for stabilizing Cr(V) at pH 5.5 and 7.5. The high redox reactivity of methyl glycosides with Cr(V) at high $[H^+]$ is attributed to the formation of $[Cr^{V}O(O,O\text{-glycoside})(OH_2)_3]^+$ species (g_{iso} 1.9716), which are not observed at pH 5.5–7.5, where only the five-coordinate bis-chelate oxochromate(V) species are observed.

Introduction

Chromium(VI) is a potential hazard both in a biological and an ecological context.[1] The observation of Cr(V) and Cr(IV) intermediates in the selective oxidation of organic substrates by Cr(VI) and their implication in the mechanism of Cr-induced cancers[2–4] has generated a considerable amount of interest in their chemistry and biochemistry.[5–8] The reduction of Cr(VI) to lower oxidation states occurs with a wide variety of biological reductants.[9–14] The major coordination sites involved in Cr binding are hydroxo, alcoholato, carboxylato, and thiolato donors groups.[1b,c,13] Five-membered, *O*-donor, chelate ligands, such as 1,2-diols and 2-hydroxy acids, are effective as non-enzymatic reductants (at low pH values) and chelates for high Cr oxidation states.[15–21] Carbohydrates constitute 5–20% (average 10%) of soil organic matter and they constitute more than 50% of dry matter in plants.[22] For this reason, it is interesting to examine the ability of saccharides and their derivatives to reduce or stabilize high oxidation states of Cr, in order to understand their potential roles in the biochemistry of Cr.

In Nature, a vast number of glycosides occur in the form of glycopyranosyl and glycofuranosyl attachments to aglycones. They are present in all plants and include a large group of phenolic glycosides, the *C*-glycosyl compounds, the thioglycosides, the physiologically important cardiac glycosides, plant pigments and many others.[23–26] In addition, the monomeric units in polysaccharides are linked by glycosidic bonds. Methyl glycosides (Gly1Me) are the simplest sugar derivatives to model the interactions of glycosides and polysaccharide with Cr(VI/V). A general pattern of reactivity has been determined for the reduction and stabilization of high oxidation states of Cr by aldoses.[27,28] However, this pattern should not apply directly to the glycoside-Cr interaction because aldoses are selectively oxidised at C^1 to yield the corresponding aldonic acid,[29–33] while in Gly1Me, the primary C^6OH group is selectively oxidised to C^6O_2H to afford the methyl glycofuranuronic acid.[34] In a previous paper, the reaction between Cr(VI) and methyl α-D-glucopyranoside (Glc1Me) and methyl α-D-mannopyranoside (Man1Me) provided the first information on the kinetics and mechanism of the reduction of chromate in acid medium.[34] However, a comparative study of a larger number of glycosides is required in order to obtain a more general reactivity pattern.

† Electronic supplementary information (ESI) available: typical kinetics plots showing the initial absorbance enhancement (Fig. 1S); effect of [Gly1Me] on k_{obs} at 40 °C and 1 M $HClO_4$ (Fig. 2S); experimental and simulated X-band EPR spectra of mixtures of (a) Cys/Cr^{VI}/α- or β-Gal1Me and $[Cr^{V}O(ehba)_2]^-$/α- or β-Gal1Me (Figs. 3S and 4S); and experimental X-band EPR spectra of 1 : 100 $[Cr^{V}O(ehba)_2]^-$: β-Glc1Me mixtures (Fig. 5S). See http://www.rsc.org/suppdata/dt/b2/b204280k/

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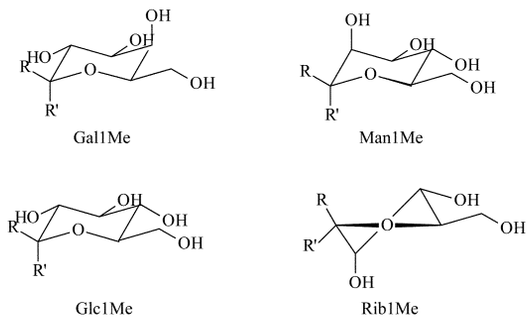
In this work, the reduction (high acid concentration) and stabilization (pH 5–7) of high Cr oxidation states by a number of methyl glycosides has been studied. The aim was to determine their relative reactivity in terms of the factors influencing the ability of the glycoside to stabilize and reduce Cr(VI) and Cr(V).



α-anomer: R = H; R' = OMe; β-anomer: R = OMe; R' = H

Experimental

Materials

Methyl α- and β-D-glucopyranoside (Sigma, 99%), methyl α- and β-D-mannopyranoside (Sigma, 98%), methyl α- and β-D-galactopyranoside (Gal1Me, Sigma, 100%), methyl α- and β-D-ribofuranoside (Rib1Me, Sigma, 100%), methyl β-D-xilofuranoside (Xil1Me, Sigma, 99%), L-cysteine (Cys, Sigma, 98%), potassium dichromate (Mallinckrodt), sodium chromate (Merck), perchloric acid (A.C.S. Baker), acrylonitrile (Aldrich, 99%), sodium hydroxide (Cicarelli, p. a.) were used without further purification. $Na[CrO(ehba)_2]\cdot H_2O$ was synthesized from 2-ethyl-2-hydroxybutanoic acid (ehba, Aldrich, 99%), and sodium dichromate (Merck) in acetone (Merck, A. R. grade) according to the literature method.[35] Water was deionized and doubly distilled from $KMnO_4$.

4-(2-Hydroxyethyl)-1-piperazineethanesulfonic acid (HEPES, Research Organics) was added to adjust the pH value of the solution to 7.5. Acetate buffer (made from Merck, AR, reagents) was used for experiments at pH 5.5. The concentrations of stock solutions of perchloric acid were determined by titration employing standard analytical methods.

CAUTION! Chromium(VI) compounds are human carcinogens, and Cr(V) complexes are mutagenic and potential carcinogens.[36] Contact with skin and inhalation must be avoided. Acrylonitrile is a carcinogen and must be handled in a well-ventilated fume hood.[37]

Reaction products

Under the conditions used in the kinetic measurements (excess of methyl glycoside over Cr(VI)), the methyl glycuronolactone (oxidation at the C^6-OH) was identified as the only reaction product by HPLC using conditions previously described.[34]

Glycoside stabilities

The stability of Gly1Me under the conditions used in the kinetic studies was checked by monitoring the optical rotation changes on a JENA polarimeter with thermostated 20-cm tubes using the sodium D line. Reactant solutions were previously thermostatted and transferred into the cell immediately after mixing. The optical rotations were recorded at different times after the preparation of the solutions and were identical to those of freshly prepared Gly1Me solutions. Specific rotations: α-Glc1Me $[\alpha]_D^{40} = +155.5° \pm 0.5°$ (c = 0.7 M, 1.0 M $HClO_4$, t = 5–1710 min); β-Glc1Me $[\alpha]_D^{40} = -32.2° \pm 0.5°$ (c = 0.31 M, 1.0 M $HClO_4$, t = 5 min to 24 h); α-Man1Me $[\alpha]_D^{40} = +70.5° \pm 0.5°$ (c = 0.56 M, 1.0 M $HClO_4$, t = 5–1440 min); α-Gal1Me $[\alpha]_D^{40} = +143° \pm 0.5°$ (c = 0.52 M, 1.0 M $HClO_4$, t = 5 min to 24 h); β-Gal1Me $[\alpha]_D^{40} = -7.4° \pm 0.5°$ (c = 0.31 M, 1.0 M $HClO_4$, t = 5 min to 24 h).

Polymerization test

The presence of free radicals in the reactions of all of the Gly1Me derivatives with Cr(VI) was tested by the acrylonitrile polymerization test. In a typical experiment, to a solution of $K_2Cr_2O_7$ ((4.00 ± 0.07) × 10^{-6} mol) and Gly1Me ((8.002 ± 0.009) × 10^{-4} mol) in 1.00 M $HClO_4$ (4.00 mL) was added acrylonitrile (1.00 mL) in 1.00 M $HClO_4$ (1 : 1) at 40 °C. After a few minutes, a white precipitate appeared. Control experiments (without $K_2Cr_2O_7$ or reductant present) did not show the formation of a precipitate. Possible reactions of Cr(V) and Cr(IV) with acrylonitrile were tested with $Na[Cr^{V}O(ehba)_2]$ and $[Cr^{IV}O(ehbaH)_2]$; the latter was generated in solution following the described method.[38] No precipitation occurred on mixing the Cr(V) or Cr(IV) complexes with acrylonitrile under the same conditions used in the Cr(VI) + glycoside reactions.

Spectrophotometric measurements

Kinetic measurements were performed by monitoring absorbance changes using a Jasco V-530 spectrophotometer with a fully thermostatted cell compartment (±0.2 °C). The reactions were followed under pseudo-first-order conditions, using at least a 300-fold molar excess of glycoside over Cr(VI). Reactant solutions were previously thermostatted and transferred into a 1-cm pathlength cell immediately after mixing. Experiments were performed at 40 °C unless otherwise stated.

The disappearance of Cr(VI) was followed at 350 nm until at least 80% of Cr(VI) was consumed. In the kinetic measurements, the concentrations of Cr and $HClO_4$ were kept constant at 6.0 × 10^{-4} M and 1.00 M, respectively, and the [β-Glc1Me], [α-Gal1Me] and [β-Gal1Me] were varied from 0.18 M to 0.48 M. In the case of β-Man1Me, α-Rib1Me and β-Rib1Me, experiments were performed at two glycoside concentrations (0.24 M and 0.36 M). The observed pseudo-first-order rate constants (k_{obs}), determined from the slopes of the linear part of plots of $\ln(A_{350})$ *vs.* time, were averages of at least three determinations and were within ±5% of each other. The k_{obs} values were determined at various $[Cr(VI)]_0$ (4.0–8.0 × 10^{-4} M) at constant temperature, initial glycoside concentration, acidity and ionic strength.

At the end of the reaction of Cr(VI) with Gly1Me in 1.00 M $HClO_4$ at 40 °C, two d–d bands were observed in the electronic absorption spectrum at λ_{max} = 409 nm (ε = 18.0 M^{-1} cm^{-1}) and 574 nm (ε = 15.3 M^{-1} cm^{-1}), which are similar to those observed for the $[Cr(H_2O)_6]^{3+}$ ion at the same pH value (410 nm, ε = 18.2 M^{-1} cm^{-1} and 574 nm, ε = 15.2 M^{-1} cm^{-1}). These bands are attributable to the octahedral $^4A_{2g} \longrightarrow {}^4T_{1g}$ and $^4A_{2g} \longrightarrow {}^4T_{2g}$ transitions in O_h symmetry, and are distinctive of the free Cr(III) aqua complex.[39] However, at the end of the reaction of Cr(VI) with Man1Me and Rib1Me in 1.00 M $HClO_4$ and 25 °C, two d–d bands were observed in the electronic absorption spectrum at λ_{max} = 409 and 574 nm, but with higher absorbances than those expected if the only product was $[Cr(OH_2)_6]^{3+}$, *i.e.*, for the Man1Me-Cr(VI) reaction (300 : 1 ratio; $[Cr(VI)] = 8.0 \times 10^{-3}$ M), when Cr(VI) was completely consumed (45 min), the observed Cr(III) absorption bands at 409 and 574 nm were 25 and 15% more intense than those calculated for 8.0 × 10^{-3} M $[Cr(OH_2)_6]^{3+}$. The intensity of these d–d bands decreased slowly with time and after two days approached those of the $[Cr(OH_2)_6]^{3+}$ ion, indicating the formation of a Cr(III)-sugar complex as the initial redox product, followed by its hydrolysis to $[Cr(OH_2)_6]^{3+}$.[40,41] This behavior was not observed for Gal1Me and Glc1Me, under the same reaction conditions.

EPR measurements

EPR spectra were obtained on a Bruker EMX spectrometer. The microwave frequency was generated with a Bruker 04 ER

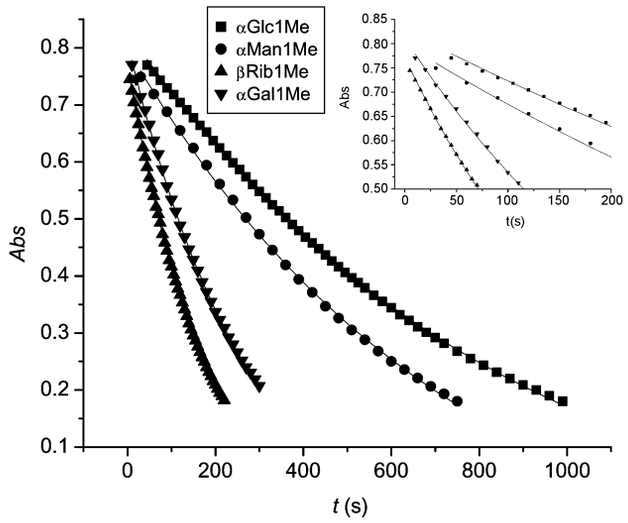


Fig. 1 Typical kinetics plots showing formation of intermediates: [Cr(VI)] = 6.0×10^{-4} M, Gly1Me : Cr(VI) = 400 : 1; [$HClO_4$] = 1.00 M, I = 1.00 M, T = 40 °C.

(9–10 GHz) and measured with a Bruker EMX 048T frequency meter. The magnetic field was measured with a Bruker EMX 035M NMR-probe gaussmeter. Reactions were carried out by addition of $Na[Cr^{V}O(ehba)_2]$ (1.0 mM), or Na_2CrO_4 (1.0 mM) + Cys (1.0 mM), to solutions of glycoside (100 mM) in acetate buffer (100 mM, pH 5.5) or HEPES buffer (100 mM, pH 7.5) at 20 ± 1 °C. Stock solutions were: $Na[Cr^{V}O(ehba)_2]$ (100 mM) in DMF; Na_2CrO_4 (100 mM) in H_2O; and Cys (100 mM) in H_2O (kept under Ar). EPR spectra were simulated using the program PEST WinSIM,[42] using 100% Lorentzian lineshapes. The spectral parameters for each Cr(V) species were consistent within all simulation, with maximum deviations in the g_{iso} values being ±0.0001 units. In the simulations, values for ^1H a_{iso} were included only where the ^1H a_{iso} value is greater than the LW (line width) of the Cr(V) species, since the signal is not significantly affected where the ^1H a_{iso} value is ≤ LW.

Results

Cr(VI) Oxidation of Gly1Me

The reduction of Cr(VI) by Gly1Me is extremely slow at pH > 1. However, the rate of the redox reaction is [H^+] dependent and at higher [H^+], Cr(VI) is rapidly consumed. Thus, the rate of reduction of Cr(VI) by methyl glycosides was compared in 1 M $HClO_4$. The kinetic traces at 350 nm show increased absorbance immediately after mixing and an initial deviation from first-order decay over very short time periods (typical kinetic curves are shown in Fig. 1).[43] However, when Cr(V) reacts faster than Cr(VI) and exists in solution at a sufficiently small concentration, changes in absorbance at 350 nm essentially reflect changes in Cr(VI) concentration (except for the short initial period) and the rate constants can be calculated from the linear part of the ln(A) *vs.* time plots, as reported for the reduction of Cr(VI) by α-Glc1Me and α-Man1Me.[34] Table 1 summarizes the values of the pseudo-first-order rate constants (k_{obs}) calculated from the slopes of the linear part of the ln(A) *vs.* time curves, for various concentrations of β-Glc1Me, α-Gal1Me and β-Gal1Me, in 1.00 M $HClO_4$. In every case, plots of k_{obs} *vs.* [Gly1Me] show a first-order dependence on [Gly1Me], and the values of the second-order rate constants (k_H) were determined from the slopes of the straight lines (Table 2). Consequently, at constant [$HClO_4$], the rate law is expressed as:

$$-d[Cr(VI)]/dt = k_{obs}\,[Cr(VI)]_T = k_H\,[Gly1Me]\,[Cr(VI)] \quad (1)$$

Table 1 Observed pseudo-first-order rate constants (k_{obs})[a] for different concentrations of Gly1Me

	$10^4\,k_{obs}/s^{-1}$		
[Gly1Me]/M	α-Gal1Me	β-Gal1Me	β-Glc1Me
0.18	33.5(3)	27.5(3)	9.1(1)
0.24	46.6(2)	35.2(3)	11.5(1)
0.30	56.8(5)	45.9(4)	13.8(1)
0.36	65.8(3)	51.7(2)	15.8(1)
0.42	77.1(9)	65.0(7)	18.6(2)
0.48	85.2(8)	70.5(5)	21.2(3)

[a] Mean values from at least three determinations, T = 40 °C; $[Cr^{VI}]_0 = 6.0 \times 10^{-4}$ M; [$HClO_4$] = 1.00 M.

Table 2 Values of the second-order rate constant (k_H) at [$HClO_4$] = 1.00 M and T = 40 °C

Gly1Me	$10^3\,k_H/M^{-1}\,s^{-1}$
α-Gal1Me	18.3(2)
β-Gal1Me	14.9(1)
α-Glc1Me	6.7(1)[a]
β-Glc1Me	4.5(1)
α-Man1Me	7.0(1)[a]
β-Man1Me	8.3(2)[b]
α-Rib1Me	15(2)[b]
β-Rib1Me	28(1)[b]

[a] Values from ref. 34. [b] Calculated as the mean of k_{obs}/[Gly1Me] for two [Gly1Me].

As expected on the basis of the rate law, $-d(\ln[Cr(VI)])/dt = k_{obs}$, where $k_{obs} = f([glycoside][H^+])$, k_{obs} was essentially constant with increasing $[Cr(VI)]_0$.

Stabilization of Cr(V) by Gly1Me at pH 5.5 and 7.5

The reduction of Cr(V) by Gly1Me is [H^+] dependent, and at the high [H^+] used in the kinetic measurements, Cr(V) intermediates rapidly yield Cr(III) and oxidized sugar redox products. At pH > 1, the redox reaction becomes slow and Cr(V) species remain in solution for longer periods of time. With this in mind, the ability of Gly1Me to stabilise Cr(V) generated in either the one-electron reduction of Cr(VI) by Cys or the ligand-exchange reactions of $[Cr^{V}O(ehba)_2]^-$ was investigated in the 5.5–7.5 pH range by EPR spectroscopy, where strong isotropic signals are observed at room temperature in X-band spectra.

At room temperature, in acetate (pH 5.5) or HEPES (pH 7.5) buffers, the reaction of chromate with Cys in the presence of a large excess Xil1Me and Glc1Me over Cr(VI) did not result in any EPR signal. Under the same conditions, the reaction of Cr(VI) with Cys (1 : 1 ratio) in the presence of 100-times excess α(β)-Gal1Me or α-Man1Me over Cr(VI), affords the Cr(V) EPR spectra shown in Fig. 2a. The best fit of spectra affords two triplets (signals were deconvoluted by fitting the spectra to Lorentzian derivatives) at g_{iso1} = 1.9802 and g_{iso2} = 1.9800/1, with the four ^{53}Cr (9.55% abundance, I = 3/2) hyperfine peaks at 16.5×10^{-4} cm^{-1} spacing (Table 3). Simulation of the spectra with a single triplet, affords spectral parameters which are not consistent for all the simulated spectra and with poorer correlation (R < 0.99) than obtained for simulations with two triplets (R > 0.995). In each case, the Cr(V) EPR signal grows much faster at pH 5.5 than at pH 7.5 as a consequence of the slower electron-transfer reaction between Cr(VI) and Cys at the higher pH value. At pH 7.5 and room temperature, the Cr^{V}-glycoside EPR signal is observed for several days.

In acetate buffer (pH 5.5) and room temperature, the ligand-exchange reaction of $[Cr^{V}O(ehba)_2]^-$ with a 100-times molar excess α(β)-Gal1Me or α-Man1Me occurs but only to a low extent. The EPR spectrum is dominated by the $[Cr^{V}O(ehba)_2]^-$

Table 3 EPR spectral parameters for Cr(v) intermediates in the reactions[a]

Cr^V species	g_{iso}	^{53}Cr $A_{iso}/10^4$ cm^{-1}	$a_H/10^4$ cm^{-1} (N)
$[CrO(O^2,O^3\text{-}\alpha Man1Me)_2]^-$	1.9802	16.5 ± 0.1	1.01 (2)
	1.9800		1.00 (2)
$[CrO(O^2,O^3\text{-}\alpha Man1Me)(ehba)]^-$	1.9792	ND	0.85 ± 0.05 (1)
	1.9790		0.60 ± 0.02 (1)
$[CrO(O^3,O^4\text{-}\alpha Gal1Me)_2]^-$	1.9802	16.5 ± 0.2	1.01 (2)
	1.9801		1.00 (2)
$[CrO(O^3,O^4\text{-}\alpha Gal1Me)(ehba)]^-$	1.9791	ND	0.87 ± 0.02 (1)
	1.9789		0.67 ± 0.03 (1)
$[CrO(O^3,O^4\text{-}\beta Gal1Me)_2]^-$	1.9802	16.5 ± 0.1	0.99 (2)
	1.9800		0.97 (2)
$[CrO(O^3,O^4\text{-}\beta Gal1Me)(ehba)]^-$	1.9791	ND	0.85 (1)
	1.9789		0.67 (1)
$[CrO(ehba)_2]^-$	1.9783	17.1 ± 0.2	—

[a] N: number of equivalent protons. ND: not determined.

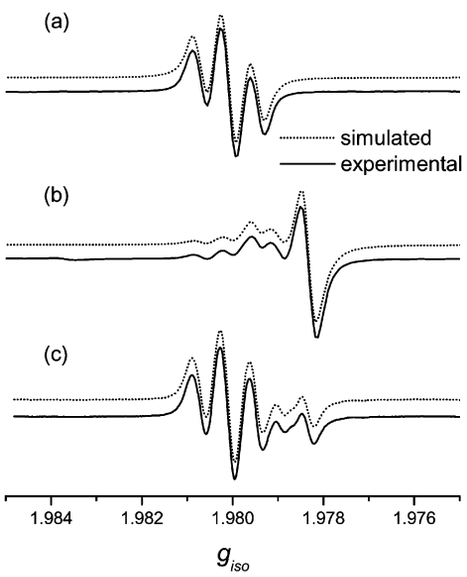


Fig. 2 Experimental and simulated X-band EPR spectra of mixtures of: (a) Cys : Cr^{VI} : α-Man1Me = 1 : 1 : 100, pH 5.5; t = 120 min; (b) $[Cr^VO(ehba)_2]^-$: α-Man1Me = 1 : 100, pH 5.5, t = 3 min; (c) $[Cr^VO(ehba)_2]^-$: α-Man1Me = 1 : 100, pH 7.5, t = 15 min, T = 25 °C, frequency ≈ 9.67 GHz, mod. ampl. = 0.2 G.

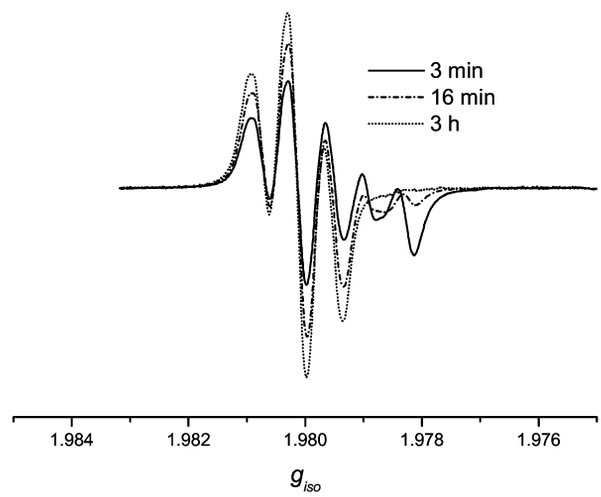


Fig. 3 Time-dependent EPR spectra of a 1 : 100 $[Cr^VO(ehba)_2]^-$: βGal1Me mixture. Conditions: pH 7.5, T = 25 °C, frequency ≈ 9.67 GHz, mod. ampl. 0.2 G.

signal at g_{iso} = 1.9783 (>50%) while the four new signals at 1.9802, 1.9800/1, 1.9791/2 and 1.9789/90, corresponding to ligand-exchange products (Figs. 2b, 3), are only minor ones, even 2 h after mixing. At room temperature and pH 7.5 (HEPES buffer), the ligand-exchange and disproportionation[8] reactions of the ehba complex take place faster and 10 min after mixing, $[Cr^VO(ehba)_2]^-$ represents less than 3% of the total Cr(v) in the reaction mixture (Fig. 2c). After 3 h, the Cr(v) triplets at g_{iso} 1.9802 and 1.9800/1 are the unique EPR features in the spectra and remain so for several days (Fig. 3).

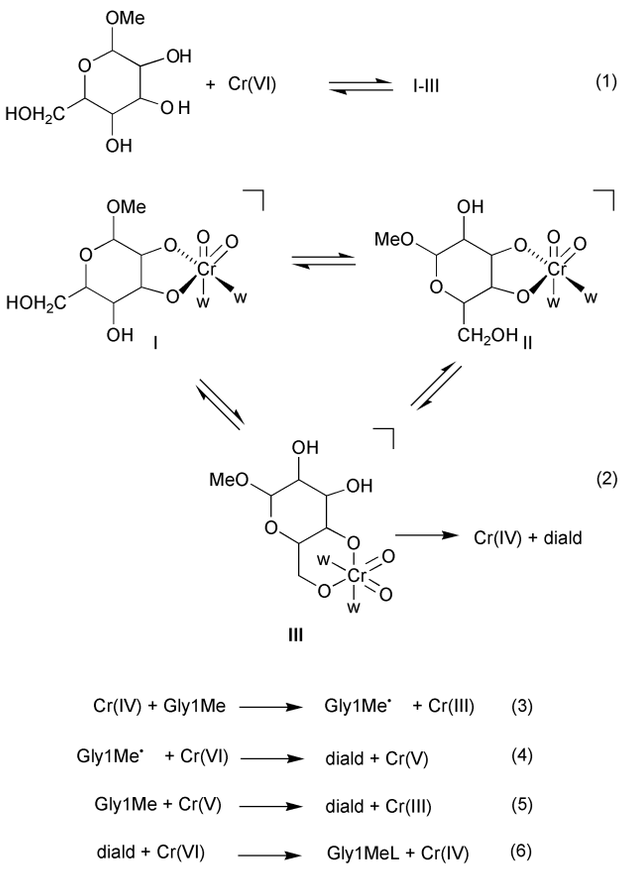
No new Cr(v) EPR signals were observed when an excess of Glc1Me or Xil1Me was added to $[Cr^VO(ehba)_2]^-$ in acetate buffer (pH 5.5). In HEPES buffer (pH 7.5), a low intensity signal at g_{iso} ≈ 1.9792 was observed a few minutes after mixing, which was replaced completely by another weak signal at g_{iso} = 1.9799 after 3 h. In this case, disproportionation[8] of $[Cr^VO(ehba)_2]^-$ is faster than the ligand-exchange reactions with Glc1Me or Xil1Me, so the maximal intensity of the new EPR signals is very low.

Discussion

Oxidation of methyl glycosides by Cr(VI)

For the substrate and acid concentrations used in this work, the oxidation of Gly1Me by Cr(VI) is a complex multi-step reaction yielding $[Cr(OH_2)_6]^{3+}$ and the methyl glycuronolactone (Gly1MeL) as the final redox products. In Scheme 1, a mechanism is proposed that takes account of: (i) the kinetic results; (ii) the polymerization of acrylonitrile added to the reaction; (iii) the increase in absorbance just after mixing of the reactants; (iv) the detection of intermediate oxochromate(v) species; and (v) the formation of Gly1MeL as the only organic reaction product.

At the $[H^+]$ and [Cr(VI)] used in the kinetic studies, Cr(VI) exists as $HCrO_4^-$,[44] and this species is proposed as the reactive form of Cr(VI) in agreement with the first-order dependence of the reaction rate on [Cr(VI)]. The chromic acid oxidations are preceded by the formation of a chromate(VI) ester complex within which the electron-transfer process takes place.[45,46] The increase in the absorbance at 350 nm immediately after mixing provides evidence for the formation of such intermediate Cr(VI) complexes. This absorbance enhancement cannot be attributed to intermediate Cr(V) species, because when Cr(V) forms and decays at a rate comparable to that of Cr(VI), the absorbance decreases more slowly than expected for a monotonic exponential decay, but it is never higher than that of the initial Cr(VI) solution (before the addition of the reductant), such as observed in the present case.[30,31,40] This absorbance enhancement is also observed a few seconds after mixing Gly1Me and Cr(VI) under conditions where the redox reaction is extremely slow, revealing that such an intermediate Cr(VI) complex is formed rapidly prior to the redox steps. The first step of the mechanism proposed in Scheme 1 is the formation of several linkage isomers of the Gly1Me–Cr(VI) monochelate (**I**, **II** and **III**) with the glycoside acting as a bidentate ligand bound to



Scheme 1 Proposed mechanism of the methyl glycoside oxidations by Cr(VI) in strongly acidic media; diald = dialdoglycoside; w = aqua ligand.

Cr(VI) *via* any pair of properly disposed hydroxyl groups. While there is no direct evidence for bidentate chelation of the sugar to Cr(VI), it is likely that this is the case, especially since Cr(VI) chelates are observed by NMR spectroscopy in the oxidations of another bidentate oxygen donor, oxalate.[47] The formation of the Gly1Me–Cr(VI) chelate is in agreement with the kinetic evidence, which suggests that a necessary step prior to oxidation of glycols is the formation of cyclic esters if these are sterically favorable.[48–51] The formation of cyclic esters has also been proposed in the Cr(VI) oxidation of the terminal alcoholic group of glycols and aminoalcohols where the two functional groups are separated by two or more C atoms.[51,52] For alditols, unfavorable electronic contributions due to the inductive effect from the OH substituents retard the redox reaction although it is partially compensated by the cyclic ester formation resulting in a rate for the redox reaction that is higher than those observed for monoalcohols.[53] Taken into account that in the redox reaction only the primary alcohol is oxidized, it is likely that the complex with the primary hydroxyl group bound to Cr(VI) (**III**) is the precursor for the slow redox steps. The slow redox step in the mechanism is proposed to take place *via* a two-electron transfer to yield Cr(IV) and the methyl dialdoglycoside (diald). After this slow redox step, Cr(IV) is predicted to react with excess glycoside to yield Cr(III) and a methyl glycoside radical in a fast step (eqn. 3 in Scheme 1). The latter is supported by the observed polymerization of acrylonitrile when it is added to the reaction mixture. The rapid reaction of the glycoside radical with Cr(VI) affords Cr(V) (eqn. 4), which can further oxidise the methyl glycoside to yield Cr(III), and the methyl dialdoglycoside (eqn. 5), which is rapidly oxidised by Cr(VI) to the methyl glycuronolactone as the final oxidation product (eqn. 6).

Previously it was shown that in the Cr(VI) oxidation of aldoses, the aldose that forms the less thermodynamically stable aldose-Cr(VI) chelate with the structure required for the intramolecular hydride transfer will be the more reactive intermediate for the intramolecular redox reaction.[27,28] Thus, the tendency in the redox reactivity of the Gly1Me toward Cr(VI) is rationalized by consideration of the relative instability of the Gly1Me–Cr(VI) chelate formed in the rapid pre-equilibrium (Scheme 1). The trend in the reactivity of the Gly1Me substrates (Fig. 4) reveals that βRib1Me reacts with Cr(VI) faster than any of the other studied glycopyranosides, a fate probably related to the furanosic form of Rib1Me. It has been proposed that the furanosic ring favors oxidation *vs.* complexation processes as a consequence of the influence of the strain-induced instability of the chromate ester on the rate of the redox reaction.[27] The tendency observed in the reactivity of the methyl glycopyranosides with Cr(VI) can be interpreted in terms of the Gly1Me–Cr(VI) chelate instability induced by the nonbonded 1,3-diaxial interactions.[29] Thus, the $O^4a : H^2a$ steric interaction in the Gal1Me stereoisomer should have the highest rate accelerating effect, followed by the $O^2a : H^4a$ interaction in Man1Me. Glc1Me, with all the ring substituents in pseudo-equatorial position, forms the most thermodynamically stable chelate and is oxidized more slowly than the other isomers.

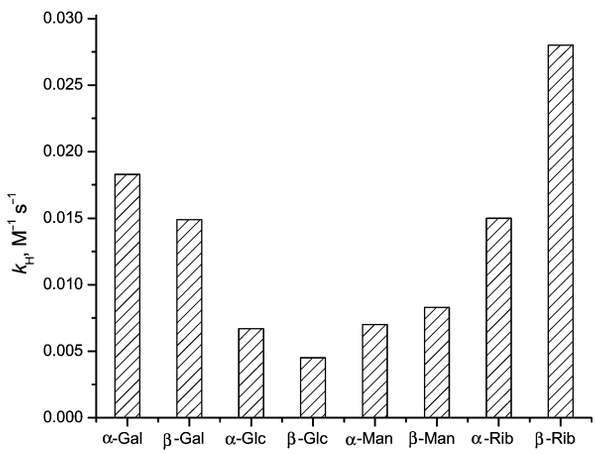
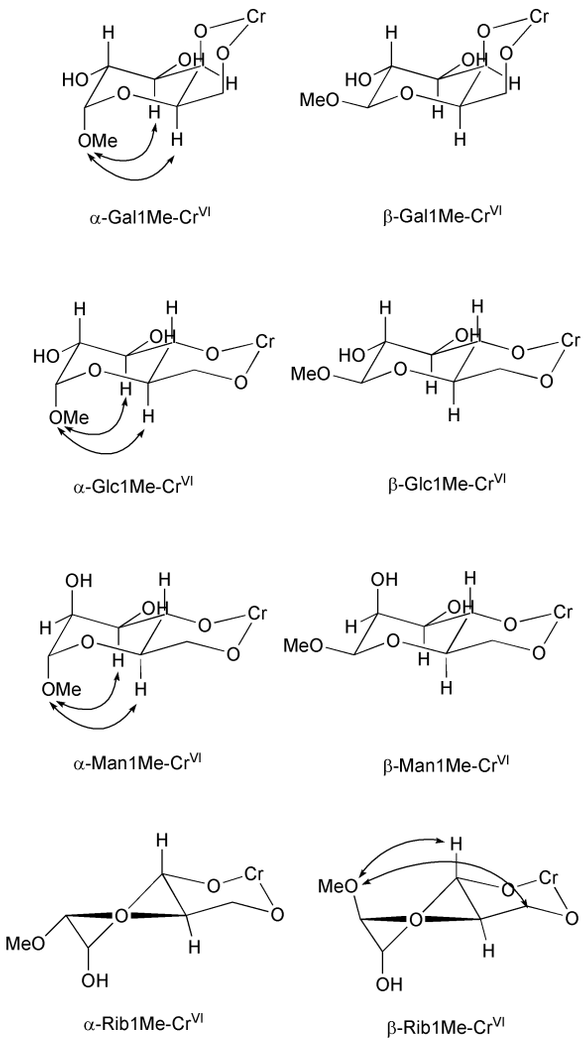


Fig. 4 Redox reactivity of Gly1Me toward Cr^{VI} in 1.00 M $HClO_4$ and $T = 40$ °C.

The relative reactivity of the α- and β-anomers of each Gly1Me is explained in terms of the relative instability of the respective Gly1Me–Cr(VI) chelates. Scheme 2 shows the additional *syn*-1,3 non-bonding interactions generated by the axial methoxy group in α- and β-O^4,O^6-Gly1Me–Cr(VI). For Gal1Me and Glc1Me, the α-anomer has two additional (Oa : Ha) steric interactions. These interactions run counter to and exceed the electronic stabilization of the axial methoxy group of this anomer,[54] so that the α-anomer is expected to be more reactive than the β-anomer, which is consistent with the results in Fig. 4. In Man1Me, the axial hydroxyl group on C^2 increases the magnitude of the anomeric effect,[54] and the result is that the β-anomer is less stable than the α-anomer. The free energy difference in favour of the αMan1Me over βMan1Me is smaller than that found for the β- over the α-anomer in Glc1Me and Gal1Me. This explains why the β-Man1Me is only slightly more reactive than α-Man1Me (Fig. 4). For Rib1Me, the magnitude of the destabilization due to *syn*-1,3 interactions is larger than the stabilization gained when the methoxy group on C^1 assumes a quasi-axial orientation, so that the β-anomer will be less stable and should react faster than the α-anomer, as is observed.

Stabilization of Cr(V) species at pH 5.5 and 7.5

The most common means of characterising Cr(V) complexes in solution is by EPR spectroscopy. The EPR spectral parameters, g_{iso} and A_{iso} values, together with the proton superhyperfine (shf) coupling, are useful in determining the binding modes of sugars to the Cr(V) center.[1b,c,27,30,31,33,55–57] An empirical relationship between the nature and number of donor groups and the



Scheme 2 1,3-Non-bonding interactions in 1,2-diolato chelates of methyl glycosides with Cr.

EPR spectral parameters of Cr(v) complexes has been established.[1c,55] Five-coordinate Cr(v) species show higher g_{iso} and lower ^{53}Cr A_{iso} values than in the corresponding six-coordinate species.[55,57–59] Thus, the assignment of the structures of new oxo-Cr(v) species in solution was made according to the isotropic EPR parameters (g_{iso} and A_{iso} values) and the superhyperfine (shf) pattern of the signal.[1b,c,57]

The EPR spectrum of a Cr(v)-diolato species of six-membered ring *cis*-diols yields a doublet, since only one proton is in the plane of the unpaired electron density of the Cr(v) ion; while the EPR spectrum of a Cr(v)-diolate species of six-membered ring *trans*-diols (with no protons lying in the ligand plane) yields a singlet.[60] Consequently, the EPR spectral multiplicity of bis-chelate Cr(v)-diolato$_2$ species formed between Cr(v) and pyranosic *cis*- and *trans*-diols will exhibit a triplet and a singlet, respectively, arising from the orientation of the ring protons of the co-ordinated diol groups with respect to the basal-plane of the complex.[1b,c,55,57,60] Based on this, the isotropic EPR parameters g_{iso} and A_{iso} were used to deduce the coordination number and the nature of the donor groups of the Cr(v) species formed in the reaction of Cr(VI) with Cys in the presence of excess glycoside and for the ligand-exchange reaction of the glycosides with $[Cr^{V}O(ehba)_2]^-$, according to a described empirical method.[1b,c,55,57–59]

The first important observation is that at pH 5.5 and 7.5 an EPR signal is observed when excess Gal1Me or Man1Me is added to a Cys : Cr(VI) equimolar mixture but not upon addition of excess Glc1Me or Xil1Me. The potential binding modes in the abovementioned cyclic glycosides to afford five-membered Cr(v) species are the 2,3-*cis*- and 3,4-*trans*-diolato moieties in Man1Me, the 2,3-*trans* and 3,4-*cis*-diolate moieties in Gal1Me, the 2,3-*trans* and 3,4-*trans*-diolato moieties in Glc1Me and the 2,3-*trans*-diolate moiety in Xil1Me. The five-membered Cr(v) chelates are the most thermodynamically favored and CrO^{3+} shows a marked preference for binding to *cis*- rather than *trans*-diol groups of cyclic diols.[27,29,34,60] Thus, the observation of the EPR signal only for Man1Me and Gal1Me reflects the expected higher ability of the *cis vs.* the *trans*-diolato for binding Cr(v). Based on this, the EPR spectral features of Man1Me- and Gal1Me–Cr(v) mixtures were interpreted in terms of Cr(v)-diolato species involving mainly binding through O^2,O^3-Man1Me and O^3,O^4-Gal1Me. The contribution to the EPR signals from other species, involving O^3,O^4-Man1Me and O^2,O^3-Gal1Me, should be small.

The low intensity EPR singlet at $g_{iso} = 1.9792$ observed at the early stages of the ligand-exchange reactions of $[Cr^{V}O(ehba)_2]^-$ with Xil1Me and Glc1Me in HEPES buffer (pH 7.5) has a g_{iso} value and the multiplicity expected for $[Cr^{V}O(ehba)(trans\text{-}O,O\text{-sugar})]^-$ ($g_{calc} = 1.9791$). After 3 h, the weak singlet at $g_{iso} = 1.9799$ is due to $[Cr^{V}O(trans\text{-}O,O\text{-Gly1Me})_2]^-$ ($g_{calc} = 1.9800$). The relatively poor ability of the *trans*-diolato groups to bind Cr(v) means the disproportionation of $[Cr^{V}O(ehba)_2]^-$ (which is fast at pH 7.5)[8] occurs much faster than the ligand-exchange reaction with the result that its EPR signal quickly disappears and only a very weak $[Cr^{V}O(O,O\text{-Gly1Me})_2]^-$ signal is still observed after 3 h. Similarly, the $[Cr^{V}O(O,O\text{-Gly1Me})_2]^-$ EPR signal is not observed for the Cr(VI)–Cys reaction in the presence of excess Xil1Me or Glc1Me because the Cys–Cr(v) redox reaction[61] is much faster than the Cr(v)–Cys ligand-exchange reactions by these glycosides. Therefore, kinetic control favors the formation of Cr(v)–diolato species of *cis*- over *trans*-diolato moieties of six-membered sugar rings, but once formed, Cr(v)-diolato of both *trans*- and *cis*-diol are inert enough toward dissociation or subsequent redox reactions to remain for long time in solution.

The two EPR triplets at $g_{iso1} = 1.9802$ and $g_{iso2} = 1.9800/1$ (A_{iso} 16.5(3) $\times 10^{-4}$ cm^{-1}, 1 : 1 g_{iso1}/g_{iso2} ratio) observed in the Cr(VI)–Cys reaction in the presence of excess α-Man1Me or α(β)-Gal1Me, at pH 5.5 or 7.5 have g_{iso} and A_{iso} values consistent with those calculated for five-coordinate oxochromate(v) complexes with four alcoholato donors ($g_{calc} = 1.9800$, $A_{calc} = 16.5 \times 10^{-4}$ cm^{-1}).[1c,55] The simulation of the shf pattern indicates that the EPR signal is a composite of two oxo-Cr(v)-*cis*-diolato$_2$ species with two (one from each chelate ring) carbinolic protons coupled to the Cr(v) electronic spin. The two components are attributed to two geometric isomers of $[Cr(O)(cis\text{-}O,O\text{-Gly1Me})_2]^-$ (**IV–VII** in Fig. 5).[60] The intensity of the Cr(v) EPR signal increases more rapidly at pH 5.5 than at pH 7.5 as a consequence of the faster formation of Cr(v) from the redox reaction between Cys and Cr(VI) at the lower pH value.[61]

At pH 7.5, the rapid ligand-exchange reactions of $[CrO(ehba)_2]^-$ with excess α-Man1Me or α(β)-Gal1Me resulted in five Cr(v) EPR signals after 15 min with $g_{iso1} = 1.9802$ (triplet), $g_{iso2} = 1.9800/1$ (triplet), $g_{iso3} = 1.9792/1$ (doublet), $g_{iso4} = 1.9789/90$ (doublet) and $g_{iso5} = 1.9783$ (singlet) (Fig. 2c). The two triplets correspond to the two geometric isomers of $[CrO(cis\text{-}O,O\text{-Gly1Me})_2]^-$ as discussed above for the Gly1Me–Cys–Cr(VI) system. The g_{iso} values and the multiplicity of the signals at $g_{iso} = 1.9791/2$ and $1.9789/90$ are consistent with those expected for the two geometric isomers of $[Cr^{V}O(ehba)(cis\text{-}O,O\text{-Gly1Me})]^-$ (**VIII–XI** in Fig. 5). The singlet at $g_{iso} = 1.9783$ is due to $[Cr^{V}O(ehba)_2]^-$. As the reaction progresses, the proportion of Cr(v)-diolate$_2$ species increases and that of $[Cr^{V}O(ehba)_2]^-$ decreases such that after 40 min, the oxo-Cr(v)-diolate$_2$ isomers are the only Cr(v) species observed in the EPR spectra in a ~1 : 1 g_{iso1}/g_{iso2} ratio (Fig. 3).

At pH 5.5, ligand-exchange reactions of $[CrO(ehba)_2]^-$ with excess α-Man1Me or α(β)-Gal1Me are not complete and after 2 h >50% of the total Cr(v) is $[CrO(ehba)_2]^-$, while oxo-Cr(v)-

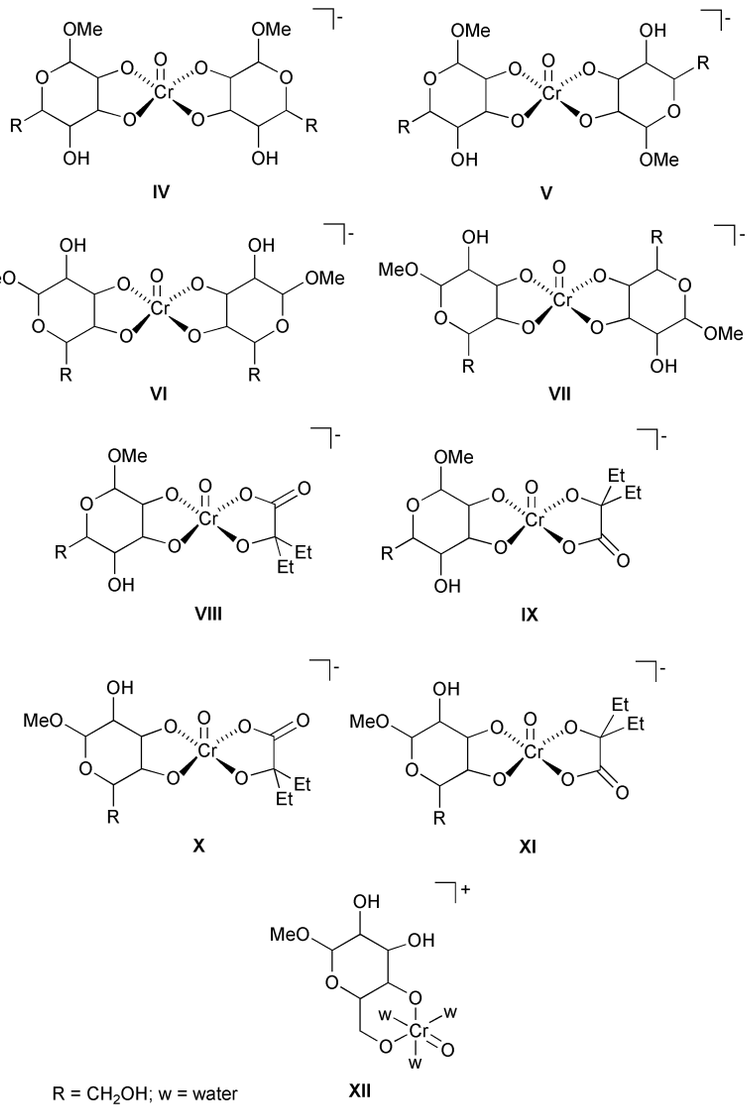


Fig. 5 Structures of Cr(v) complexes with Gly1Me ligands.

diolate$_2$ species represent ~5% of total Cr(v). This behavior is not due to the lower ability of the glycoside to bind to Cr(v) at the lower pH value, because in the Cr(VI)-Cys-Gly1Me systems the oxo-Cr(v)-diolate$_2$ is formed faster at pH 5.5 than 7.5. It is due, in part, to the lower reactivity of $[CrO(ehba)_2]^-$ toward disproportionation at lower pH values.

Under acidic conditions (1 M $HClO_4$), Gly1Me substrates reduce both Cr(VI) and Cr(v). At high $[H^+]$ complexation of Cr(v) also takes place prior to the redox paths, but the distribution of Cr(v) species is rather different than at pH ≥ 5.5. Previously, it was shown that in the reaction of α-Man1Me with Cr(VI), at pH ≤ 1 and a 10 : 1 α-Man1Me : Cr(VI) ratio, the EPR spectrum was composed of a triplet at g_{iso1} = 1.9800, a doublet at g_{iso2} = 1.9798, and three signals with no shf structure at g_{iso3} = 1.9788, g_{iso4} = 1.9746 and g_{iso5} = 1.9716.[34] The intensity of signals at g_{iso4} and g_{iso5} relative to those of the g_{iso1}, g_{iso2} and g_{iso3} signals, increased with higher $[H^+]$ and at $[H^+]$ > 0.3 M the signal at g_{iso5} was the dominant one. The species at g_{iso5} was attributed to six-coordinate $[CrO(O^3,O^4\text{-Man1Me})(OH_2)_3]^+$ (g_{calc} = 1.9724). Thus, the formation of positively charged Cr(v) monochelates are favored in acid medium and low glycoside : Cr(VI) ratios, while the formation of anionic Cr(v) bis-chelates are favored at the higher pH and larger glycoside : Cr(VI) ratios. The observation of six-coordinate $[Cr^VO(O^3,O^4\text{-Man1Me})(OH_2)_3]^+$ only at high $[H^+]$, together with the selectivity of the redox reaction (oxidation of the Gly1Me occurs selectively at the primary alcohol group), suggest that **XII** could be a six-coordinate Cr(v) monochelate precursor of the intramolecular electron-transfer reaction step in the redox reactions.

Conclusions

The *cis*-diolato group in α-Man1Me or α(β)-Gal1Me is responsible for the high chelating ability of these glycosides toward Cr(v). Intramolecular competition between *cis*- and *trans*-diolato groups favors Cr(v) binding at the *cis*-diolato moiety on the basis of the low reactivity shown by Xil1Me or Glc1Me with only *trans*-diolato groups. However, the decomposition rate of the oxo-Cr(v)-diolate$_2$ chelates is independent of the *cis*/*trans* relative configuration of the diolate since, once formed, both the oxo-Cr(v)-*cis*-diolate$_2$ and the oxo-Cr(v)-*trans*-diolate$_2$ remain in solution for a long time. The chelating behavior is essentially independent of the configuration of the pyranosic ring, since α-Man1Me and α-Gal1Me behave in the same way in either their abilities to trap Cr(v) generated by the reaction of Cys with Cr(VI), or from the ligand-exchange reactions of $[CrO(ehba)_2]^-$. In addition, the α and β isomers of Gal1Me yield the oxo-Cr(v)-diolate$_2$ geometric isomers (**VI**, **VII**) in similar final proportions and reaction times.

At pH 5.5–7.5, redox reactions between Cr(v) and Gly1Me are extremely slow as once the bis-chelates are formed they are long-lived. A different reactivity pattern is observed at high $[H^+]$, where intramolecular electron-transfer processes take place. In general, the redox rate increases with increasing

non-bonding 1,3-diaxial interactions in the Gly1Me–Cr(VI/V) monochelate precursor of the redox steps, and the high reactivity of furanosic Gly1Me in redox reactions with Cr(VI/V) is attributable to the additional strain-induced instability of the chromate ester, which lowers the energy of the transition state with respect to those of the reactants.

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References

1 (*a*) C. B. Klein, in *Toxicology of Metals*, ed. L. W. Chang, CRC-Lewis Publishers, New York, 1996, pp. 205–220; (*b*) R. Codd, C. T. Dillon, A. Levina and P. A. Lay, *Coord. Chem. Rev.*, 2001, **216–217**, 533; (*c*) A. Levina, R. Codd, C. T. Dillon and P. A. Lay, *Prog. Inorg. Chem.*, submitted.
2 S. A. Katz and H. Salem, *The Biological and Environmental Chemistry of Chromium*, VCH, New York, 1994, pp. 65–119.
3 X. Shi, A. Chiu, C. T. Chen, B. Halliwell, V. Castranova and V. Vallyathan, *J. Toxicol. Environ. Health, Part B*, 1999, 87.
4 M. Costa, *Crit. Rev. Toxicol.*, 1997, **27**, 431.
5 E. S. Gould, *Coord. Chem. Rev.*, 1994, **135/136**, 651.
6 L. Zhang and P. A. Lay, *J. Am. Chem. Soc.*, 1996, **118**, 12624.
7 D. K. Geiger, *Coord. Chem. Rev.*, 1997, **164**, 261.
8 A. Levina, P. A. Lay and N. E. Dixon, *Inorg. Chem.*, 2000, **39**, 385.
9 D. H. Stearns and K. E. Wetterhahn, *Chem. Res. Toxicol.*, 1994, **7**, 219.
10 K. E. Wetterhahn Jannette, *J. Am. Chem. Soc.*, 1982, **104**, 874.
11 P. O'Brien, J. Barrett and F. Swanson, *Inorg. Chim. Acta*, 1985, **108**, L19.
12 P. A. Lay and A. Levina, *J. Am. Chem. Soc.*, 1998, **120**, 6704.
13 M. Ciéslak-Golonka, *Polyhedron*, 1996, **15**, 3667.
14 L. Zhang and P. A. Lay, *Aust. J. Chem.*, 2000, **53**, 7.
15 M. Krumpolc, B. G. de Boer and J. Rocek, *J. Am. Chem. Soc.*, 1978, **100**, 145.
16 S. L. Brauer and K. E. Wetterhahn, *J. Am. Chem. Soc.*, 1991, **113**, 3001.
17 D. M. L. Goodgame, P. B. Hayman and D. E. Hathaway, *Polyhedron*, 1982, **1**, 497.
18 D. M. L. Goodgame and A. M. Joy, *J. Inorg. Biochem.*, 1986, **26**, 219.
19 R. Codd, P. A. Lay and A. Levina, *Inorg. Chem.*, 1997, **36**, 5440.
20 R. P. Farrell, P. A. Lay, A. Levina, I. A. Maxwell, R. Bramley, S. Brumby and J. Ji, *Inorg. Chem.*, 1998, **37**, 3159.
21 R. Codd, A. Levina, L. Zhang, T. W. Hambley and P. A. Lay, *Inorg. Chem.*, 2000, **39**, 990.
22 N. Mehta, P. Dubach and H. Deuel, *Adv. Carbohydr. Chem. Biochem.*, 1958, **16**, 335.
23 A. Das and C. V. N. Rao, *Tappi*, 1964, **47**, 339.
24 A. Das and C. V. N. Rao, *Aust. J. Chem.*, 1965, **18**, 845.
25 T. Reichstein, *Angew. Chem., Int. Ed. Engl.*, 1962, **1**, 572.
26 R. J. McIlroy, *The Plant Glycosides*, Longmans and Green, New York, 1951.
27 S. Signorella, V. Daier, S. García, R. Cargnello, J. C. González, M. Rizzotto and L. F. Sala, *Carbohydr. Res.*, 1999, **316**, 14.
28 S. Signorella, S. García and L. F. Sala, *J. Chem. Educ.*, 1999, **76**, 405.
29 S. Signorella, R. Lafarga, V. Daier and L. F. Sala, *Carbohydr. Res.*, 2000, **324/2**, 127.
30 V. Daier, S. Signorella, M. Rizzotto, M. I. Frascaroli, C. Palopoli, C. Brondino, J. M. Salas-Peregrin and L. F. Sala, *Can. J. Chem.*, 1999, **77**, 57.
31 L. F. Sala, S. Signorella, M. Rizzotto, M. I. Frascaroli and F. Gandolfo, *Can. J. Chem.*, 1992, **70**, 2046.
32 M. Rizzotto, S. Signorella, M. I. Frascaroli, V. Daier and L. F. Sala, *J. Carbohydr. Chem.*, 1995, **14**, 45.
33 S. Signorella, M. Rizzotto, V. Daier, M. I. Frascaroli, C. Palopoli, D. Martino, A. Bousseckson and L. F. Sala, *J. Chem. Soc., Dalton Trans.*, 1996, 1607.
34 S. Signorella, M. I. Frascaroli, S. García, M. Santoro, J. C. González, C. Palopoli, N. Casado and L. F. Sala, *J. Chem. Soc., Dalton Trans.*, 2000, 1617.
35 M. Krumpolc and J. Rocek, *Inorg. Synth.*, 1980, **20**, 63.
36 International Agency for Research on Cancer (IARC). *IARC Monogr. Eval. Carcinog. Risk Chem. Hum. Suppl.*, 1987, vol. 7, 165.
37 R. Feldam, in *Occupational and Environmental Neurotoxicology*, Lippincott-Raven, Philadelphia, 1999, pp. 337–338.
38 M. C. Ghosh and E. S. Gould, *Inorg. Chem.*, 1991, **30**, 491.
39 A. B. P. Lever, *Inorganic Electronic Spectroscopy*, 2nd edn., Elsevier, Amsterdam, 1984, p. 419.
40 S. Signorella, M. Santoro, C. Palopoli, C. Brondino, J. M. Salas-Peregrin, M. Quirós and J. F. Sala, *Polyhedron*, 1998, **17**, 2739.
41 S. Signorella, M. Santoro, A. Frutos, G. Escandar, J. M. Salas Peregrin, V. Moreno, M. González Sierra and L. F. Sala, *J. Inorg. Biochem.*, 1999, **73**, 93.
42 WinSIM EPR calculations for MS-Windows,version 0.96, National Institute of Environmental Health Sciences, Research Triangle Park, NC, USA, 1995.
43 G. Haight, G. Jwisich, M. Kelso and P. Merril, *Inorg. Chem.*, 1985, **24**, 2740.
44 N. E. Brasch, D. A. Buckingham, A. B. Evans and C. R. Clark, *J. Am. Chem. Soc.*, 1996, **118**, 7969.
45 J. K. Beattie and G. P. Haight, in *Inorganic Reaction Mechanisms. Part II*, ed. J. O. Edwards, Wiley, New York, 1972.
46 M. Mitewa and P. Bontchev, *Coord. Chem. Rev.*, 1985, **61**, 241.
47 R. P. Farrell, PhD Thesis, University of Sydney, 1993.
48 J. Rocek and F. H. Westheimer, *J. Am. Chem. Soc.*, 1962, **84**, 2241.
49 Y. W. Chang and F. H. Westheimer, *J. Am. Chem. Soc.*, 1960, **82**, 1401.
50 H. Kwart, J. A. Ford, Jr. and G. C. Corey, *J. Am. Chem. Soc.*, 1962, **84**, 1252.
51 K. G. Srinivasan and J. Rocek, *J. Am. Chem. Soc.*, 1978, **100**, 2789.
52 C. Palopoli, S. Signorella and L. F. Sala, *New J. Chem.*, 1997, **21**, 343.
53 V. Roldán, V. Daier, B. Goodman, M. Santoro, J. C. González, N. Calisto, S. Signorella and L. F. Sala, *Helv. Chim. Acta*, 2000, **83**, 3211.
54 J. F. Stoddart, *Stereochemistry of Carbohydrates*, Wiley-Interscience, New York, 1971, pp. 67–72.
55 G. Barr-David, M. Charara, R. Codd, R. P. Farrell, J. A. Irwin, P. A. Lay, R. Bramley, S. Brumby, J.-Y. Ji and G. R. Hanson, *J. Chem. Soc., Faraday Trans.*, 1995, **91**, 1207.
56 M. Branca, A. Dessí, H. Kozlowski, G. Micera and J. Swiatek, *J. Inorg. Biochem.*, 1990, **39**, 217.
57 J. A. Irwin, PhD Thesis, University of Sydney, 1998.
58 R. Bramley, J.-Y. Ji, R. J. Judd and P. A. Lay, *Inorg. Chem.*, 1990, **29**, 3089.
59 R. P. Farrell, R. J. Judd, P. A. Lay, R. Bramley and J.-Y. Ji, *Inorg. Chem.*, 1989, **28**, 3401.
60 R. Codd and P. A. Lay, *J. Am. Chem. Soc.*, 1999, **121**, 7864.
61 P. A. Lay and A. Levina, *Inorg. Chem.*, 1996, **35**, 7709.