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On the Interaction of Vanadium Species with the Monoisoamyl Ester of *Meso*-2,3-Dimercaptosuccinic Acid

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Abstract The interaction of the VO^{2+} cation with the monoisoamyl ester of *meso*-2,3-dimercaptosuccinic acid (MiADMSA) was investigated by electron absorption spectroscopy in aqueous solutions at different pH values. The spectral behavior, complemented with a spectrophotometric titration, shows the generation of a $[VO(MiADMSA)_2]^{4-}$ complex in which the oxocation interacts with two pairs of deprotonated –SH groups of the ester. Besides, MiADMSA rapidly reduces VO_3^- to VO^{2+}, which might be chelated by an excess of the ester, and also produces relatively rapid reduction of V_2O_5 suspensions at pH=6.5. The results of this study suggest that MiADMSA might be a potentially useful detoxification agent for vanadium.

Keywords Monoisoamyl DMSA · Oxovanadium(IV) · Vanadium(V) · Electronic spectra · Vanadium detoxification

Introduction

Environmental contamination by vanadium has dramatically increased during the last decades, especially in the most developed countries, due to the widespread use of fossil fuels, many of which liberate finely particulate vanadium pentoxide to the atmosphere during combustion [1]. Therefore, also owing to the emerging interest in the pharmacological effects of some of its compounds [2], the toxicology and detoxification of vanadium constitute areas of increasing research interest. The older literature about vanadium toxicology has been reviewed in the classical work of Faulkner-Hudson [3], and we have

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analyzed the most relevant aspects of its detoxification some years ago [4], emphasizing the relevance of chelation therapies and related chemical detoxification procedures.

Chelation therapy occupies a central place in modern medicine and pharmacology, as continuous studies with laboratory animals and extensive clinical experience demonstrate that acute or chronic intoxications with a variety of metals can be considerably improved by administration of a suitable chelating agent [5–9]. These agents are organic compounds capable of linking metal ions to form complex ring-like structures called "chelates." These compounds must show low toxicity, and their affinity should be higher for the toxic metal ion than for essential biometals. On the other hand, an ideal chelating agent must have good water solubility, the possibility of oral administration, ability to penetrate cell membranes, and must produce the rapid excretion of the chelated metal species [5, 7]. An important number of chelating agents has been developed and were successfully employed to overcome different types of intoxication. This is the case of 2,3-dimercaptopropanol (British anti-Lewisite), D-penicillamine, and different salts of EDTA and similar polyaminopolycarboxylic acids [5–9].

In recent years, two new and very promising chelating agents have been introduced into the medical praxis. They are *meso*-2,3-dimercapto succinic acid (DMSA) and 2,3-dimercapto-1-propanesulfonic acid (DMPS) [6, 7, 10–12]. Both drugs, which are very stable and show low toxicity and side effects, are available as tablets for oral administration [6, 10, 13–15].

Although DMSA is an effective and potent chelating drug for the treatment of a variety of metal intoxications, because of its hydrophilic and lipophobic properties, it is unable to pass through cell membranes, and as a consequence, it cannot capture intracellulary deposited toxic species. Thus, a large number of esters of DMSA have been synthesized to overcome this problem and to enhance tissue uptake. In order to make the compounds more lipophilic, the carbon chain length of the parent DMSA was increased by controlled esterification with different alcohols (methyl, ethyl, propyl, isopropyl, butyl, isobutyl, pentyl, isopentyl, and hexyl) [12, 16]. Most of these esters have been assayed for the treatment of metal poisoning, and it has been reported that they have a better potential for the mobilization of toxic metals than DMSA [12]. Among these new chelators, the monisoamyl ester of DMSA (MiADMSA, Fig. 1) has been found particularly effective for the detoxification of cadmium, mercury, and arsenic [12, 17–20].

As part of our studies on different aspects of vanadium biochemistry [21], toxicity [22], and detoxification [1, 4], we are especially interested in the investigation and development of new possible detoxification agents for this element. Recently, we have shown that both DMSA [23] and DMPS [24] may be potentially useful detoxification agents for vanadium.

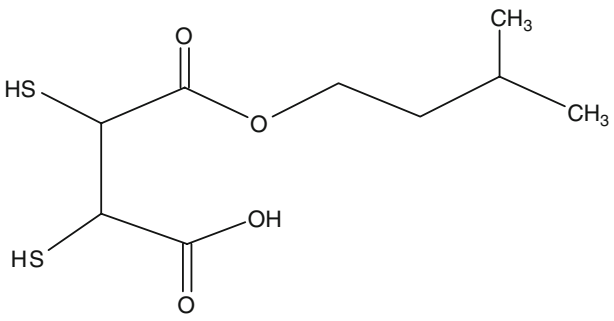

Fig. 1 Schematic structure of the monoisoamyl ester of 2,3-*meso*-dimercaptosuccinic acid (MiADMSA)

In this paper, we present the results of our studies on the interaction of different vanadium species with MiADMSA.

Materials and Methods

Commercially available DMSA from Sigma, isoamyl alcohol from Baker, $NaVO_3$ from Aldrich, and $VOCl_2$ (50% solution) from Carlo Erba were used as supplied. Pure and finely dispersed V_2O_5 was obtained by slow heating of NH_4VO_3 (Carlo Erba) up to 450°C in a muffle furnace in air [25]. The ester MiADMSA was prepared by the controlled esterification of DMSA with isoamyl alcohol in acidic (HCl) medium [16]. The product was characterized by its melting point and by ^{1}H and ^{13}C NMR spectroscopy. All the experiments were performed under a nitrogen atmosphere to avoid oxidation phenomena. The electronic spectra were measured with a Hewlett-Packard 8452 diode-array spectrophotometer, using 10-mm quartz cells. Metal and ligand solutions were mixed in different stoichiometries, and the desired pH values were adjusted either with diluted HCl or NaOH solutions.

Results and Discussion

It is well known that, in the presence of molecules containing thiol groups such as L-cysteine, reduced glutathione, or dithiothreitol, vanadium(V) is usually redox labile, i.e., it is reduced to VO^{2+} [2]. This behavior was also confirmed in the case of DMSA and DMPS [23, 24]. Preliminary qualitative experiments clearly showed that also MiADMSA can reduce vanadium(V) to vanadium(IV). Therefore, we have first examined the characteristics of the interaction of the reduced species [oxovanadium(IV), VO^{2+}] with MiADMSA to attain an insight into the form in which this species can be stabilized by an excess of this reducing ligand.

MiADMSA is not very soluble in water at non-physiological pH values. We found that the lowest pH value at which VO^{2+}/MiADMSA interactions could be studied is about 6.3. This interaction was investigated working at a 10:1 chelator/metal ratio, dissolving 1 mmol of MiADMSA in 20 mL of distilled water and adding 0.1 mmol of $VOCl_2$. The solutions remained clear in the pH range between 6.3 and 11.0, without any sign of hydrolytic phenomena. The obtained solutions showed a characteristic electronic spectrum (Fig. 2) with two defined bands at 630 nm (ε=60 $mol^{-1}cm^{-1}$) and 558 nm (ε=54 $mol^{-1}cm^{-1}$) and a strong charge transfer band at the blue, whose maximum could not be determined with certainty. The position of the observed electronic transitions were comparable to that found in the VO^{2+}/DMSA [23] and VO^{2+}/DMPS [24] systems, as well as in the complexes of the VO^{2+} cation generated with dithiothreitol [26] and with a series of bis(*N*,*N*-disubstituted) dithiocarbamates [27]. In all these cases, VOL_2 species were generated and coordination takes place through two pairs of deprotonated –SH groups. Although precise information on the p*K* values of MiADMSA is not available, the comparison with similar molecules suggests that the first deprotonation occurs at the carboxylic acid group, at rather acidic pH values, whereas the two –SH moieties are deprotonated at higher pH values. This means that, under the investigated experimental conditions, the ester is present as a trinegatively charged anion. On the other hand, the involvement of the carboxylate group in bonding can be totally excluded on the basis of the electronic spectra, as VO^{2+}/carboxylate interactions would generate absorption bands above 700 nm [28].

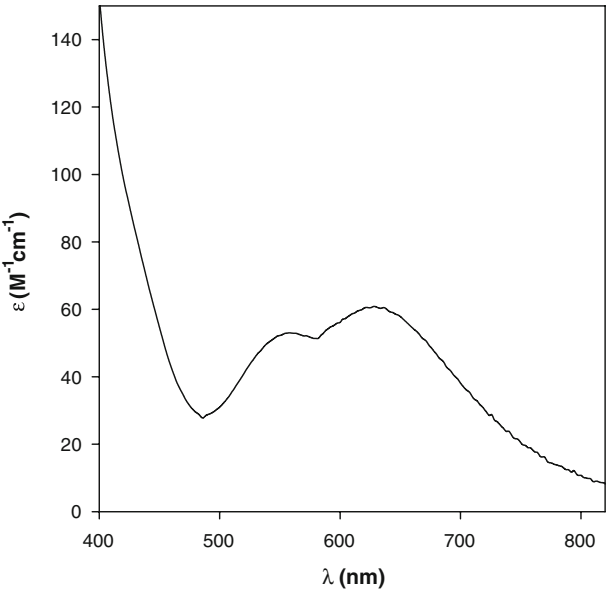

Fig. 2 Electronic absorption spectrum of an aqueous solution of MiADMSA/VO^{2+} at a 10:1 ratio (pH7.0) under a N_2 atmosphere

In order to establish the stoichiometry of the VO^{2+}/MiADMSA complex, a spectrophotometric titration [29] was undertaken. A series of solutions with ligand to metal ratios, [*L*]/[*M*], ranging from 0.5 to 10 were prepared and their pH value fixed at 6.5. Absorbance measurements were performed at 630 nm. The plot of absorbance against [*L*]/[*M*] values is shown in Fig. 3. The inflection point confirms the formation of a 2:1 MiADMSA/VO^{2+} complex. This fact, together with the spectral characteristics commented above, confirms the formation of a complex with a $[VO(MiADMSA)_2]^{4-}$ stoichiometry and the pair of deprotonated –SH groups of each ligand coordinating to the oxocation.

To verify the reducing action of MiADMSA over vanadium(V) species, we have also investigated its interaction with $NaVO_3$ and V_2O_5. For the experiments with vanadate, a solution containing a 10:1 MiADMSA/$NaVO_3$ ratio (1 mmol of the ligand and 0.1 mmol of $NaVO_3$, in 20 mL of water) was prepared, and a very rapid reduction of vanadate(V) to oxovanadium(IV) was observed. The evolution of the reaction was monitored at pH6.5 as a function of time. Already after 2 min, the same spectrum as that shown in Fig. 2 was obtained, confirming the formation of the same $[VO(MiADMSA)_2]^{4-}$ species as described above.

In the case of V_2O_5, 1 mmol of MiADMSA was added to a suspension of 0.05 mmol of the oxide in 20 mL of water, with the pH value adjusted to 6.5. After a few minutes, the reduction reaction became evident and the spectrophotometric monitoring of the reaction showed a continuous increment in absorbance with time. As shown in Fig. 4, after 35 min, the spectral pattern of the $[VO(MiADMSA)_2]^{4-}$ complex was clearly visible, and it became totally defined at about 50 min, together with the total dissolution of the V_2O_5 suspension. Interestingly, in the previously investigated systems, with DMSA and DMPS, this reaction was appreciably slower, and finally, part of the suspended pentoxide remained undissolved [23, 24].

The usefulness of the use of antioxidants along or in conjunction with chelation therapy has yet not been extensively investigated [12]. We have previously explored the therapeutic efficacy of both antioxidant and chelating agents for the removal of vanadium(V) [1, 23, 24].

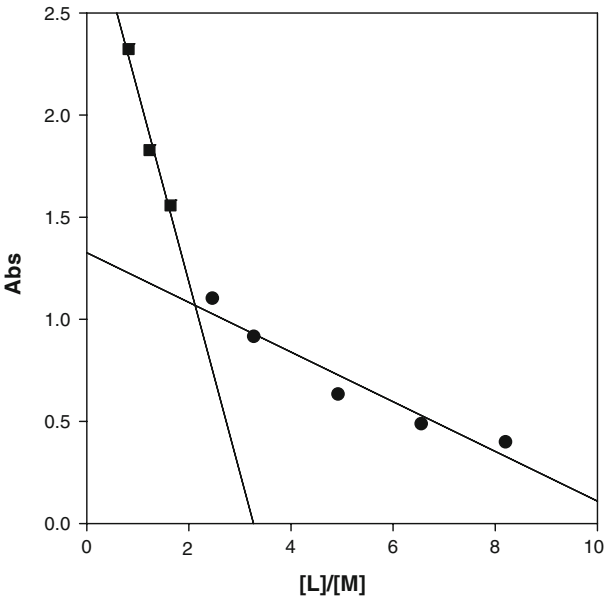

Fig. 3 Spectrophotometric titration of VO^{2+} with MiADMSA at pH=6.5 under a N_2 atmosphere and at λ=630 nm. $[L]/[M]$=[MiADMSA]/[VO^{2+}] (*Abs* absorbance in arbitrary units)

In this work, it has been demonstrated that MiADMSA is a better reducing agent than DMSA and DMPS and is able to reduce vanadate(V), and even fine suspensions of V_2O_5, to VO^{2+}. Besides, the reduced VO^{2+} species is also rapidly complexed by an excess of this chelating agent forming a 2:1 MiADMSA/VO^{2+} complex, which is stable, at least, at physiological pH values (in the pH range between 6.3 and 11.0). Therefore, MiADMSA appears as a very promising and interesting detoxification agent for vanadium(V), which merits to be further explored, for example, with laboratory animals as a next step.

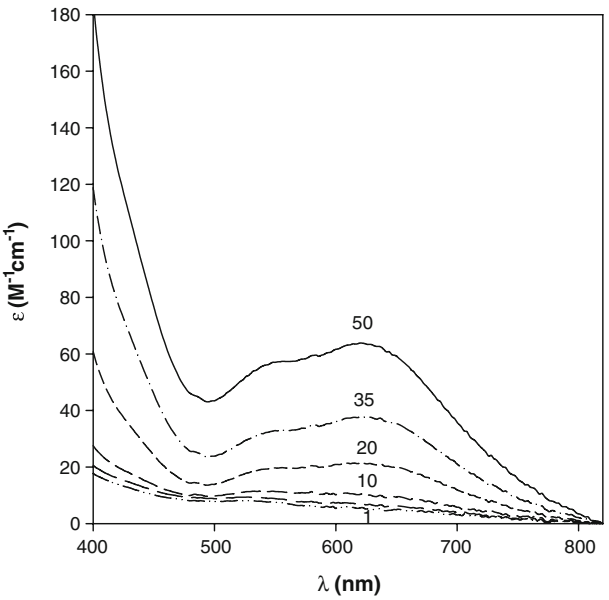

Fig. 4 Electronic absorption spectra of a suspension of 0.05 mM of V_2O_5 mixed with 1 mM of MiADMSA in water (pH6.5) under a N_2 atmosphere, measured at different times after mixing (1, 5, 10, 20, 35, and 50 min)

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References

1. Baran EJ (2008) Vanadium detoxification: chemical and biochemical aspects. Chem Biodivers 5:1475–1484
2. Rehder D (2008) Bioinorganic vanadium chemistry. Wiley, Chichester
3. Faulkner-Hudson TG (1964) Vanadium: toxicology and biological significance. Elsevier, Amsterdam
4. Baran EJ (1998) Vanadium detoxification. In: Nriagu JO (ed) Vanadium in the environment. Part II: Health effects. Wiley, New York, pp 317–345
5. Taylor DM, Williams DR (1995) Trace element medicine and chelation therapy. Royal Society of Chemistry, Cambridge
6. Andersen O (1999) Principles and recent developments in chelation treatment of metal intoxication. Chem Rev 99:2683–2710
7. Andersen O (2004) Chemical and biological considerations in the treatment of metal intoxications by chelating agents. Mini Rev Med Chem 4:11–21
8. Baran EJ (1995) Química bioinorgánica, chapter 19. McGraw Hill Interamericana de España S.A, Madrid
9. Blanusa M, Varnai VM, Piasek M, Kostial K (2005) Chelators as antidotes of metal toxicity. Therapeutic and experimental aspects. Curr Med Chem 12:2771–2794
10. Aposhian HV, Maiorino RM, González-Ramírez D, Zuniga-Charles M, Xu Z, Hurlbut KH, Junco-Munoz M, Dart RC, Aposhian MM (1995) Mobilization of heavy metals by newer, therapeutically useful chelating agents. Toxicol 97:23–38
11. Domingo JL (1998) Developmental toxicity of metal chelating agents. Reproduct Toxicol 12:499–510
12. Kalia K, Flora SJS (2005) Strategies for the safe and effective therapeutic measures for chronic arsenic and lead poisoning. J Ocupp Health 47:1–21
13. Chisholm JJ Jr, Thomas DJ (1985) Use of 2, 3-dimercaptopropane-1- sulfonate in treatment of lead poisoning in children. J Pharmacol Exper Ther 235:665–669
14. Bosque MA, Domingo JL, Paternain JL, Llobet JM, Corbella J (1990) Evaluation of the developmental toxicity of 2, 3-dimercapto-1- propanesulfonate (DMPS) in mice. Effect on mineral metabolism. Toxicol 62:311–320
15. Aposhian MM, Maiorino RM, Xu Z, Aposhian HV (1996) Sodium 2, 3- dimercapto-1-propanesulfonate (DMPS) treatment does not redistribute lead or mercury to the brain of rats. Toxicol 109:49–55
16. Jones MM, Singh PK, Gale GR, Smith AB, Atkins LM (1992) Cadmium mobilization in vivo by intraperitoneal administration of monoalkyl esters of meso-2, 3-dimercaptosuccinic acid in the mouse. Pharmacol Toxicol 70:336–343
17. Gale GR, Smith AB, Jones MM, Singh PK (1993) Meso-2, 3-dimercaptosuccinic acid monoalkyl esters: effects on mercury levels in mice. Toxicol 81:49–56
18. Xu C, Holscher MA, Jones MM, Singh PK (1995) Effects of monoisoamyl meso-2, 3-dimercaptosuccinate on the pathology of acute cadmium intoxication. Toxicol Environ Health 45:261–277
19. Flora SJS, Bhadauria S, Kannan GM, Singh N (2007) Arsenic induced oxidative stress and role of antioxidant supplementation during chelation: A review. J Environm Biol 28:333–347
20. Mishra D, Flora SJS (2008) Quercitin administration during chelation therapy protects arsenic-induced oxidative stress in mice. Biol Trace Elem Res 122:137–147
21. Baran EJ (2003) Model studies related to vanadium biochemistry: recent advances and perspectives. J Braz Chem Soc 14:878–888
22. Baran V, Baran EJ (2002) Contaminación atmosférica por vanadio: Importancia de su monitoreo y control. Anales Acad Nac Cs Ex Fís Nat 54:171–177
23. Williams PAM, Baran EJ (2006) On the interaction of vanadium species with meso-2, 3-dimercaptosuccinic acid. Biol Trace Elem Res 109:189–194
24. Williams PAM, Baran EJ (2008) Vanadium detoxification: On the interaction of oxovanadium(IV) and other vanadium species with 2, 3-dimercapto-1-propanesulfonate. J Inorg Biochem 102:1195–1198
25. Duval C (1963) Inorganic thermogravimetric analysis. Elsevier, Amsterdam
26. Williams PAM, Barrio DA, Etcheverry SB (1999) Interactions of vanadyl(IV) with dithiothreitol and thioglycolic acid. Their action on alkaline phosphatase activity. J Inorg Biochem 75:99–104
27. McCormick B (1968) Structure and spectra of dithiocarbamate complexes of oxovanadium(IV). Inorg Chem 7:1965–1970
28. Baran EJ (2001) Review: Spectroscopic studies of oxovanadium coordination compounds. J Coord Chem 54:215–238
29. Connors KA (1981) Binding constants. Wiley, New York