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A comparison between the chelating properties of some dihydroxamic acids, desferrioxamine B and acetohydroxamic acid

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Abstract

The complexes of hexanedioic acid bis(3-hydroxycarbamoyl-methyl)amide (Dha1) and hexanedioic acid bis(3-hydroxycarbamoyl-propyl)amide (Dha2) with cobalt(II), nickel(II), copper(II), zinc(II), iron(III), calcium(II) and magnesium(II) have been studied by pH-metric and spectrophotometric methods. All the complexes formed with Dha2 are soluble in water, but a very insoluble complex is formed in the copper(II)-Dha1 system. Besides the 1:1 species complexes with 2:3 stoichiometry are also formed in the cobalt(II)-, nickel(II)-, zinc(II)- and iron(III)-containing systems. Dha2 generally forms more stable complexes than those of Dha1 (e.g. log β values for their iron(III) 1:1 complexes are 17.9 and 19.1, respectively). A comparison of the data with those on the complexes of a simple monohydroxamate, (acetohydroxamate, Aha), nonano-dihydroxamate (Dha3) and the natural trihydroxamate-based siderophore, desferrioxamine B (DFB) revealed that the stability sequence of the complexes is generally: $\mathrm{DFB}>\mathrm{Dha2}\geq\mathrm{Dha3}\sim\mathrm{Dha1}>\mathrm{Aha}$. The shorter but more flexible connecting chain of Dha3 results in the ca. same stability of complexes of Dha1 and Dha3. The above sequence, however, did not hold for copper(II) allowing the coordination of at most two hydroxamates and for calcium(II). In this latter case, Dha2, containing the longest connecting chain, formed the most stable complexes. © 1999 Elsevier Science Ltd. All rights reserved.

Keywords: Dihydroxamic acids; Acetohydroxamic acid; Desferrioxamine B; Transition metal complexes; Ca(II) and Mg(II) complexes

1. Introduction

Microbial siderophores are relatively low molecular weight compounds synthesized in order to solubilize and transport iron(III) into the cells in the necessary concentrations [1,2]. Most of the hydroxamate-type siderophores contain three chelating groups (-CONHO^-) and complete the octahedral coordination sphere of the iron(III) in their 1:1 complexes. In some cases, however, microbes synthesize dihydroxamate-type siderophores which form very stable dinuclear species, $[Fe_2L_3]$, with the iron(III) [3–5]. (The log β value for the iron(III)-rhodotoluric acid complex, for example, is ca. 62 [4].) Synthetic dihydroxamate-based siderophore analogues have also been prepared and their metal complexes, primarily the iron(III) complexes, have been studied [6–10]. These earlier studies focused mainly on the effects of the connecting chain length on the stabilities and stoichiometries of the complexes. Interactions between different metal ions and primary dihydroxamic acid derivatives, ($\text{HONOC-(CH}_2)_n\text{-CONHOH}$; $n=$ 4–8) were studied by Evers et al. [8], who found that the stabilities of the 1:1 complexes increased in parallel with increasing chain length from $n=4$ to $n=7$ and then levelled out at $n=7$. Both equilibrium and kinetic studies on the iron(III) complexes of *N*-methyl-substituted dihydroxamic acids (n varying from 2 to 8) demonstrated the formation of the dimeric 1:1 species $[Fe_2L_2]^{2+}$ if $n<6$, monomeric $[FeL]^+$ if $n>6$ and the simultaneous existence of monomeric and dimeric species if $n=6$ [9,10]. As dihydroxamate-based ligands can not fully satisfy the six-coordinate geometry of iron(III), there is a strong tendency to the formation of the dimer $[Fe_2L_3]$. Its structure (triply bridged or singly bridged) has been presumed to be determined by the value of n [10].

In contrast, a significant dependence of the stability constants on the chain length was not found for the bulky *N*-isopropyl-substituted derivatives, where the formation of triply bridged $[Fe_2L_3]$ and double bridged $[Fe_2L_2]$ dinuclear species was observed [6].

The reactions between molybdenum(VI) and numerous dihydroxamic acids (including Dha1 and Dha2) were investigated in a recent study [11]. Dha1 and Dha2 were

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found to form water-soluble complexes with molybdenum(VI), whereas many other dihydroxamic acids with shorter chain lengths furnished precipitates. Polynuclear species were presumed to precipitate in these cases. It was also established that Dha2 formed more stable complexes than those of Dha1 [11].

The biologically important calcium(II) and magnesium(II) ions in general form unstable complexes. This may be the reason why their complexes with hydroxamic acids have been studied in only a very few reports [8].

In the present work, the complexes formed in aqueous solution between hexanedioic acid bis(3-hydroxycarbamoyl-methyl)amide (Dha1), or hexanedioic acid bis(3-hydroxycarbamoyl-propyl)amide (Dha2) and cobalt(II), nickel(II), copper(II), zinc(II), iron(III), calcium(II) or magnesium(II) were characterized. The results were compared with those on some other ligands, acetohydroxamic acid (Aha), nonano-dihydroxamic acid (Dha3) and the natural trihydroxamic acid desferrioxamine B (DFB), and the effects of the peptide moieties in the connecting chains, the lengths of these chains and the ionic radii of the metals on the stabilities and stoichiometries of the complexes were evaluated. Some of the metal ion-DFB, metal ion-Dha3 and metal ion-Aha systems had been studied previously [8,12–14], and these earlier results were utilized in the present work. In some other cases, the data on the model systems were determined in the present work.

2. Experimental

2.1. Chemicals

Dha1 and Dha2 were prepared as previously described [11]. Aha was purchased from Sigma. DFB was produced by CIBA Geigy. A standard procedure was used to prepare Dha3 [15]. The levels of purity of the ligands and the concentrations of the ligand stock solutions were determined by Gran's method [16].

The metal ion stock solutions were prepared from $CoCl_2 \cdot 6H_2O$, $NiCl_2 \cdot 6H_2O$, $CuCl_2 \cdot 2H_2O$, $FeCl_3 \cdot 6H_2O$ or $CaCl_2$ (Reanal) (to prevent hydrolysis of the metal ion, the iron(III) stock solution contained a known concentration of HCl (0.1 mol/dm^3)), whereas ZnO and MgO (Reanal) were dissolved in a known amount of HCl. The concentrations of the cobalt(II), nickel(II), copper(II), zinc(II) and iron(III) stock solutions were determined gravimetrically via precipitation of the quinolin-8-olates. EDTA was used as titrant to determine the concentrations of the calcium(II) and magnesium(II) stock solutions.

2.2. Potentiometric and spectroscopic studies

The pH-metric and spectrophotometric measurements were carried out at an ionic strength of 0.2 mol/dm^3 (KCl). Carbonate-free KOH solutions of known concentrations (ca. 0.2 mol/dm^3) were used as titrant.

The pH-metric titrations were performed throughout the approximate pH range 2.0–11.0 (or below precipitation) on samples of 10.00 or 25.00 cm^3.

For a higher extent of complex formation a metal ion excess was applied only in the case of calcium(II), because parallel titrations of samples containing only strong acid or strong acid plus calcium-chloride (1.36×10^{-2} mol/dm^3), or strong acid plus magnesium-chloride (1.81×10^{-2} mol/dm^3) demonstrated that no measurable hydrolysis occured in the presence of calcium(II) in the studied pH-range, but there was observable hydrolysis with magnesium(II) above ca. pH 8.

In the metal–ligand systems, the ligand concentrations were varied in the range of 8×10^{-3}–2×10^{-3} mol/dm^3; the metal–ligand ratios were in general in the range of 1:1–1:5, but in the case of calcium(II) it was increased to 20:1. Measurements were made with samples at five or six different ratios. The pH-metric measurements were made with a Radiometer pHM84 instrument equipped with a CMAWL/5 Russel combined electrode. The titrant was added from a Metrohm 715 Dosimat autoburette. The electrode system was calibrated by the method of Irving et al. [17] so that the pH-meter readings could be converted into hydrogen ion concentrations.

The UV–visible measurements on the iron(III)-Dha1, iron(III)-Dha2, iron(III)-Dha3 and iron(III)-DFB systems were carried out in two sets. Individual samples were prepared in the first series, in which the 0.2 mol/dm^3 KCl was partially or completely replaced by HCl. The pH values, varying in the range 0.7–1.3, were calculated from the HCl content. In the second set, titrations were made on samples containing the iron(III) ion at 5×10^{-4} mol/dm^3 or 4×10^{-4} mol/dm^3. The metal ion to ligand ratios were 1:2.5, 1:3.75 and 1:5, and the spectra were recorded at 15–20 different pH values. A sample of iron(III)-Aha containing the iron(III) in 4×10^{-4} mol/dm^3, the Aha in 3×10^{-3} mol/dm^3 was also titrated. A HP8453 spectrophotometer was used to record the spectra in the region 300–600 nm.

Both pH-metric and spectrophotometric results were utilized to establish the stoichiometry of the species and to calculate the stability constants of the iron(III) complexes and the pH-metric experimental data in all the other cases. The calculations were performed with the computer program PSEQUAD [18]. Literature data on the iron(III) hydroxo complexes were included in the models [19]. In the pH-regions where the experimental findings indicated the possibility of hydrolysis (a continuous decrease of the pH or the formation of a precipitate), calculations were not performed. The parameters fitted by the PSEQUAD program were the volumes of titrant in the case of the pH-metric data, and the absorbances of spectrophotometric titration curves. Accepted fittings were below 1.10^{-2} in the former cases, and below ca. 2.10^{-2} in the latter.

3. Results and discussion

3.1. Protonation of the ligands

The formulae of Dha1, Dha2 and the model ligands, Aha, Dha3 and DFB, are depicted in Scheme 1. The dissociation constants of the ligands are listed in Table 1. The values determined in the present work are in very good agreement with those published earlier in our former works [11–13] or by others [8].

3.2. Metal complexation of Dha1 and Dha2

Representative pH-metric titration curves for different metal ion-Dha2 systems are shown in Fig. 1. Those for the Dha1-containing systems are similar (except for copper(II) -Dha1, where precipitation hindered the measurements), and they are therefore not presented here.

In Fig. 1, the significant differences in the pH effects caused by the complex formation with various metal ions clearly reveal the great differences in stability of the complexes. Whereas the complex formation with iron(III) starts at very low pH, there is no interaction with calcium(II) up to pH ca. 7. (In the cases of calcium(II) and magnesium(II), titration curves for 1:1 metal to ligand ratio, where noticeable differences could already be observed between the curves for the free ligand and for the metal-containing systems; see Fig. 1).

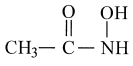
Table 1
Dissociation constants (p*K*) for the ligands (t=25°C; I=0.2 mol/dm^3 KCl)

	pK_1	pK_2	pK_3	pK_4
Aha[a]	9.27(1)			
Dha1	8.48(1)	9.25(1)		
Dha2	8.86(1)	9.64(1)		
Dha3[b]	9.00(2)	9.84(2)		
DFB[c]	8.30(1)	9.00(1)	9.46(1)	10.84(3)

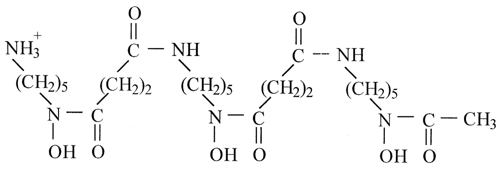
[a] Ref. [8]; pK_1=9.36; t=25°, I=0.10 mol/dm^3 (KCl).

[b] Ref. [8]; pK_1=9.02, pK_2=9.65; t=25°, I=0.10 mol/dm^3 (KCl).

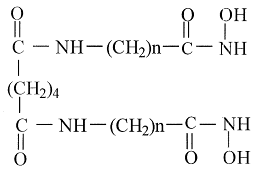
[c] Ref. [8]; pK_1=8.32, pK_2=8.96, pK_3=9.55, pK_4=10.79; t=25°, I= 0.10 mol/dm^3 (KCl).



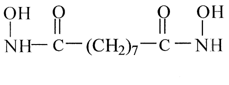
Acetohydroxamic acid (Aha)



Desferrioxamine B (DFB)



n = 1 Hexanedioic acid bis(3-hydroxycarbamoyl-methyl)amide (Dha1)
n = 3 Hexanedioic acid bis(3-hydroxycarbamoyl-propyl)amide (Dha2)



Nonano-dihydroxamic acid (Dha3)

Scheme 1.

As the pH-metric method can not be used in the strongly acidic pH range (below ca. pH 2), spectrophotometric measurements were also performed in the case of the iron(III)-containing systems. Well-known characteristic charge-transfer absorptions allow determination of the stoichiometry and stability constants of the complexes [2]. Each added hydroxamate group results in characteristic changes in both λ_{max} and the absorption. The coordination of one hydroxamate chelate results in λ_{max}~510 nm and ϵ~1000 mol^{-1} cm^{-1}, two coordinated hydroxamates lead to λ_{max}~470 nm and ϵ~1800 mol^{-1} cm^{-1}, while complexes with three coordinated hydroxamates have λ_{max}=420–430 nm, ϵ~2600–2800 mol^{-1} cm^{-1}.

Some of the spectrophotometric spectra recorded for iron(III)-Dha1 samples in the pH ranges 0.7–1.3 (a) and ca. 2–10 (b) are presented in Fig. 2.

When the above mentioned characteristics are considered, the following conclusions can be drawn from the spectra: Two hydroxamate chelates are already coordinated to the iron(III) at below pH 1 (see Fig. 2a), while the coordination of the third hydroxamate starts only at ca. pH 3. The pH-range in which the three-chelated complex predominates is quite narrow (5.5–6.5). Above this pH-range the absorbance begins to decrease indicating a decrease in the number of coordinated hydroxamates, most probably because of hydrolysis. The behaviour of Dha2 exhibits many similarities (which is why these spectra are not shown here), but also some differences, e.g. the pH-region, in which only the three-chelated complex exists is somewhat wider (ca. pH 5–8.0).

Both pH-metric and spectrophotometric experimental data were used to calculate the stability constants in the pH regions where no decrease in absorbance was observed (pH≤6.5 in the case of Dha1, and pH≤8.0 in the case of Dha2). The best fits were found on the assumption of the existence of $[FeL]^+$ and $[Fe_2L_3]$ for both ligands. The monoprotonated, monochelated species $[FeLH]^{2+}$ was formed below the studied pH region (see Fig. 2a), and thus

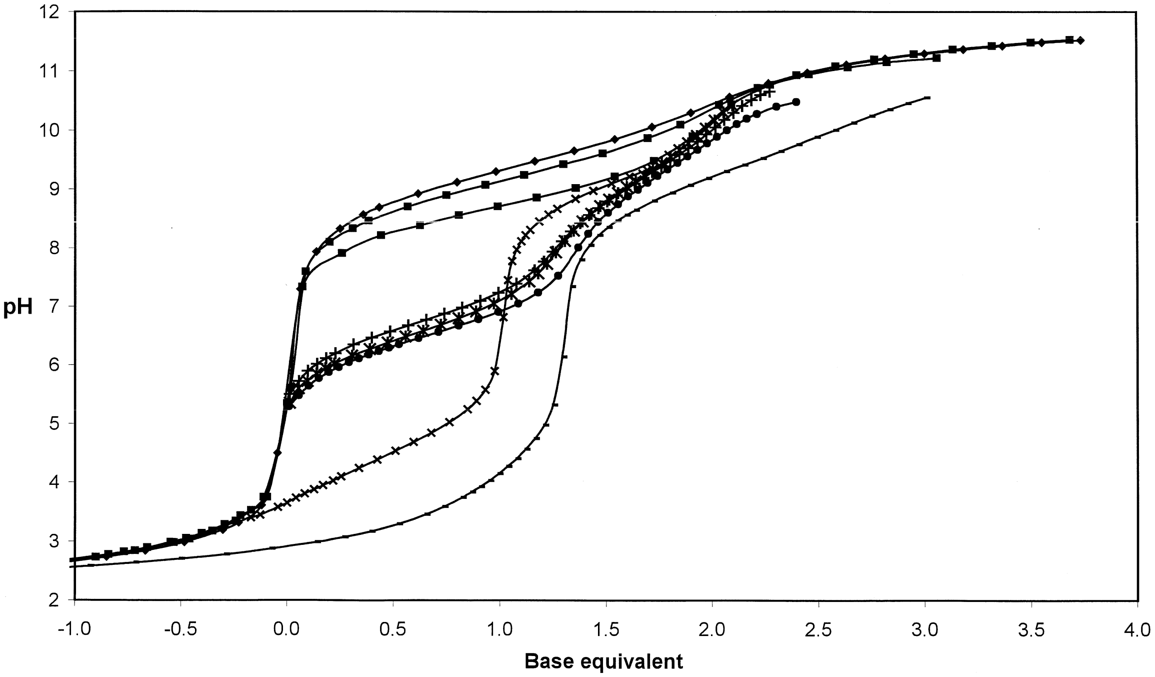


Fig. 1. pH-metric titration curves for the Dha2 (♦) and calcium(II) (■), magnesium(II) (▲), cobalt(II) (+), nickel(II) (*), zinc(II) (●), copper(II) (×), iron(III) (−) Dha2 systems. $c_{\text{lig}}=5\times10^{-3}$ mol/dm^3. Metal ion to ligand ratios can be found in the parenthesis. Ca(II) (1:1), Mg(II) (1:1); Co(II) (1:1.5); Ni(II) (1:1.5); Zn(II) (1:1.5); Cu(II) (1:1.5); Fe(III) (1:2.4). Negative base equivalent values mean acid excess.

the corresponding stability constants could not be determined.

The copper(II)-Dha1 complexes are insoluble in water which strongly suggests the formation of polynuclear complexes. The complexes formed with Dha2, however, are water-soluble and the pH-metric titration curves could be fitted on the assumption of $[\text{CuLH}]^+$ and [CuL]. Complexes with stoichiometries $[\text{MLH}]^+$, [ML] and $[\text{M}_2\text{L}_3]^{2-}$ were necessary to fit the titration curves for the cobalt(II)-, nickel(II)- or zinc(II)-Dha1 and Dha2 systems.

Because of the low stabilities of the calcium(II)- and magnesium(II)-dihydroxamate complexes, the pH-effects of their formation are very small. This is well demonstrated in Fig. 1. The small, but repeatable pH-effects, however, allow calculation of the stability constants of the 1:1 complexes in both the calcium(II)- and the magnesium(II)-containing systems.

All the equilibrium models and calculated stability constants are listed in Table 2.

Protonated complexes (1:1:1) with dihydroxamic acids may be formed if one of the chelating moieties remains in protonated non-coordinated form. These species are formed in the measurable pH-range for all of the studied metals except iron(III). It is known from previous studies [6–10] (see Introduction) that, depending on the length and character of the connecting chain, the two hydroxamate chelates of any dihydroxamic acid coordinate either to the same metal ion (1:1 species) or to different ones (2:2 species). In all the present cases, the inclusion of the 1:1 species in the models gave the best fits, supporting the possibility of the formation of the mononuclear 1:1 species, [ML], containing the two hydroxamate chelates coordinated to the same metal ion. It appears most likely

Table 2

Models and stability constants (log β) for the complexes formed in iron(III)-, copper(II)-nickel(II)-, zinc(II)-, cobalt(II)-, calcium(II)- and magnesium(II)-Dha1 and Dha2 systems

$$\beta_{\text{M}_p\text{L}_q\text{H}_r}=\frac{[\text{M}_p\text{L}_q\text{H}_r]}{[\text{M}]^p[\text{L}]^q[\text{H}]^r}$$

($t=25$°C; $I=0.2$ mol/dm^3 KCl)

Metal ion	Ionic radius (pm)	$M_pL_qH_r$			Dha1	Dha2
		p	q	r		
Fe(III)	55	1	1	0	17.9(1)	19.1(1)
		2	3	0	51.6(1)	55.6(1)
Cu(II)	73	1	1	1	prec.	17.28(9)
		1	1	0		12.83(1)
Ni(II)	69	1	1	1	13.89(2)	14.71(2)
		1	1	0	7.44(1)	8.33(4)
		2	3	0	20.94(5)	22.66(4)
Zn(II)	74	1	1	1	13.98(4)	14.62(3)
		1	1	0	7.73(5)	8.33(5)
		2	3	0	22.01(7)	23.01(5)
Co(II)	74	1	1	1	13.61(3)	14.43(4)
		1	1	0	6.87(5)	7.89(3)
		2	3	0	19.0(4)	21.91(5)
Ca(II)	100	1	1	1	11.08(5)	11.87(7)
		1	1	0	2.68(2)	3.09(2)
Mg(II)	72	1	1	1	11.72(7)	12.51(4)
		1	1	0	3.46(2)	4.11(1)
		1	1	−1	−8.21(9)	−7.65(5)

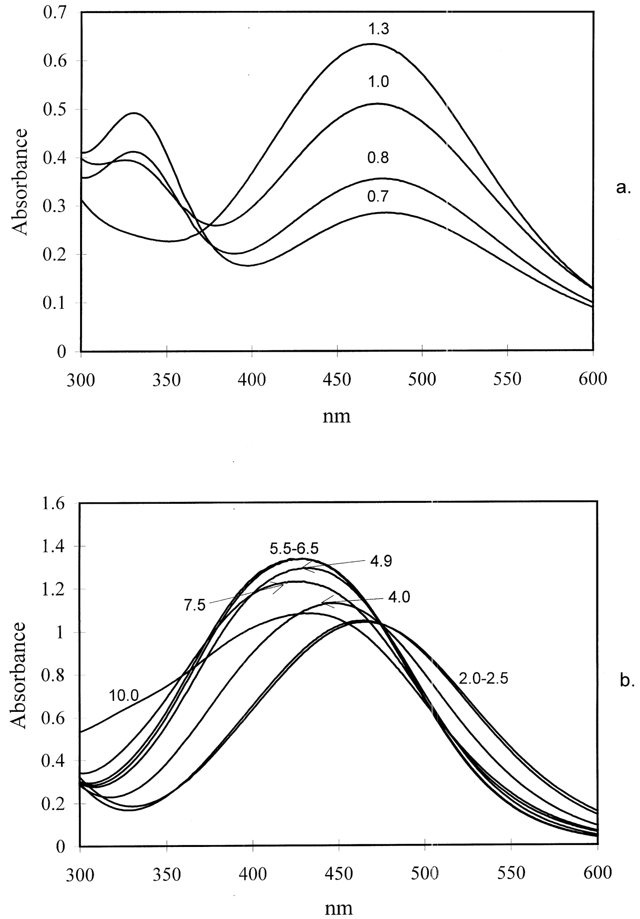


Fig. 2. UV–vis spectra recorded for iron(III)-Dha1 system in the pH-ranges 0.7–1.3 (a) and 2.0–10.0 (b). ($c_{\mathrm{Fe(III)}} = 5\times10^{-4}$ mol/dm^3, $c_{\mathrm{Dha1}} = 2.5\times10^{-3}$ mol/dm^3).

that two [ML] are joined by the third ligand in the species [M_2L_3].

All the stability constants for the complexes with Dha2 in Table 2 are somewhat larger than those for the complexes with Dha1. The differences are to some extent due to the differences in basicity of the coordinating donors (see Table 1), but other factors, e.g. differences in steric hindrances, may also be important.

3.3. Metal complexation with Aha, Dha3 and DFB

The stability constants for most of the nickel(II), zinc(II), copper(II) and iron(III) complexes of these ligands have already been determined either by the present authors [12–14] or by Evers et al. [8]. However, as the constants for the iron(III) complexes with Dha3 and with DFB were determined earlier under different conditions, we have determined them again in the present work. The complex-forming behaviour of cobalt(II) towards the dihydroxamate-based ligands was found to be very similar to that of nickel(II) and zinc(II). For this reason, we did not study the cobalt(II)-Aha, -Dha3 and -DFB systems. In the cases of calcium(II) and magnesium(II), we had to study the systems with all these three ligands. The experimental results relating to the magnesium(II)-DFB and especially the calcium(II)-DFB system are particularly interesting. The pH effects of the complex formation in these systems are even less than those in the case of Dha2 (see Fig. 1).

The overall stability constants for the complexes with Aha, Dha3, DFB are reported in Table 3.

3.4. A comparison between the chelating properties of the different ligands

Different possibilities are available to compare the metal-binding abilities of Dha1, Dha2, Aha, Dha3 and

Table 3

Overall stability constants (logβ) for the complexes formed in iron(III)-, copper(II)-, nickel(II)-, zinc(II)-, calcium(II)- and magnesium(II)-Aha, -Dha3 and -DFB systems[a] ($t = 25$°C; $I = 0.2$ mol/dm^3 KCl)

Metal ion	$M_pL_qH_r$			Aha	Dha3	DFB
	p	q	r			
Fe(III)	1	1	0	11.09[a]	19.13(5)	30.4(1)
	1	2	0	20.69[a]		
	1	3	0	28.80[a]		
	1	1	1			41.01(1)
	2	3	0		56.0(1)	
	1	1	2			42.4(1)
Cu(II)	1	1	3			36.99[d]
	1	1	2			33.10[d]
	1	1	1			23.98[d]
	1	1	0	7.89[b]	13.60[d]	13.73[d]
	2	1	1			32.09[d]
	1	2	0	14.06[a]		
	1	2	−1	4.44[a]		
Ni(II)	1	1	3			33.20[d]
	1	1	2			27.66[d]
	1	1	1		14.89[c]	19.71[d]
	1	1	0	5.15[b]	8.92[c]	8.89[d]
	1	2	0	9.18[b]		
	1	3	0	11.68[b]		
	1	1	−1	−4.35[b]		
Zn(II)	1	1	3			33.40[d]
	1	1	2			28.17[d]
	1	1	1			20.40[d]
	1	1	0	5.18[b]		10.36[d]
	1	2	0	9.45[b]		
	1	3	0	11.57[b]		
	1	1	−1	−3.40[b]		
Ca(II)	1	1	2			22.41(4)
	1	1	1		12.07(5)	13.25(3)
	1	1	0	2.45(1)	2.78(2)	3.03(3)
Mg(II)	1	1	2			23.85(3)
	1	1	1		12.69(8)	14.66(3)
	1	1	0	2.96(3)	4.15(3)	2.8(1)
	1	1	−1	−7.22(9)		

[a] Standard deviations in parenthesis are shown if only the values were determinated in the present work.

[b] Ref. [12].

[c] Ref. [15].

[d] Ref [8]; $t = 25$°C, $I = 0.1$ mol/dm^3, ($NaNO_3$).

[e] Ref. [13].

Table 4
Derived stability constants calculated for iron(III)-, copper(II)-, nickel(II)-, zinc(II)-, calcium(II)- and magnesium(II)-Aha, -Dha1, -Dha2, -Dha3 and -DFB complexes

Ligand	Equilibrium processes	Fe(III)	Cu(II)	Ni(II)	Zn(II)	Ca(II)	Mg(II)
Aha	M+L	11.09	7.89	5.15	5.18	2.45	2.96
	ML+L	20.69	14.06	9.18	9.45		
	ML_2+L	28.80		11.68	11.57		
Dha1	M+LH			4.64	4.73	1.83	2.47
	M+L	17.9		7.44	7.73	2.68	3.46
	2ML+L	15.8		6.06	6.55		
Dha2	M+LH		7.64	5.07	4.98	2.23	2.87
	M+L	19.1	12.83	8.33	8.33	3.09	4.11
	2ML+L	17.4		6.00	6.35		
Dha3	M+LH			5.05		2.23	2.85
	M+L	19.13	13.60	8.92		2.78	4.15
	2ML+L	17.74					
DFB	M+LH_3		7.69	3.90	4.10		
	M+LH_2	22.1	12.80	7.36	7.87	2.11	3.55
	M+LH	30.2	13.14	8.87	9.56	2.41	3.82
	M+L	30.4	13.73	8.89	10.36	3.03	2.80

DFB. For instance, we can compare the stoichiometries and stabilities of the complexes and the pH-ranges of their formation. As Tables 2 and 3 show, monochelated species were not formed in measurable concentrations in the iron(III)-dihydroxamate or-DFB systems. We could not determine stability constants for the [FeLH] species with dihydroxamic acids or for [$FeLH_3$] with DFB. The monochelated iron(III) complex was formed only with Aha. On the other hand, many protonated complexes were formed between the different dihydroxamic acids or DFB and copper(II), nickel(II), zinc(II), cobalt(II), calcium(II) or magnesium(II). In these cases, the overall stability constants include the corresponding protonation constants. To compare the stabilities of the hydroxamate chelates, different derived constants were calculated. These values and the related equilibrium processes are shown in Table 4.

The derived constants in Table 4 reveal that the Dha1 complexes display the lowest stability in most cases. The differences in stability are the most significant in the case of iron(III). The stability sequence of iron(III) complexes is well demonstrated in Fig. 3, which depicts the pH-dependence of the absorbance at 420 nm. The values were taken from the spectra of samples with the same hydroxamate group/iron(III) concentration ratio.

Fig. 3 illustrates the constant absorbances for iron(III)-DFB in the pH range 2–10. The calculated value of $E°$ (2650 M^{-1} cm^{-1}) indicates that exclusively the three-chelated complex ([FeL]) exists in the studied pH range for DFB. The three-chelated species ([Fe_2L_3]), however, predominates only in the pH range 5.0–8.0 for Dha2, and in an even narrower pH range (5.5–6.5) with Dha1. If we compare the pH ranges where exclusively [Fe_2L_3] is formed with Dha3 (having the shortest, but flexible connecting chain) and with Dha1, the former (5.5–8.0) is significantly wider. The difference cannot be explained solely by the differences in basicity of the coordinating donor atoms in the two ligands. There may be another contribution from the fact that the presence of two peptide groups in the relatively short connecting chain of Dha1 makes the chain quite inflexible. Below the pH ranges where exclusively the three-chelated species are formed, processes of protonation of the chelating moieties compete with metal complex formation, while above these pH ranges hydrolytic processes may occur. Formation of the three-chelated species with Aha ([FeL_3]) becomes predominant only at ca. pH 6 and the decrease in absorbance (indicating a decrease in the number of coordinated hydroxamates) starts only at ca. pH 8.

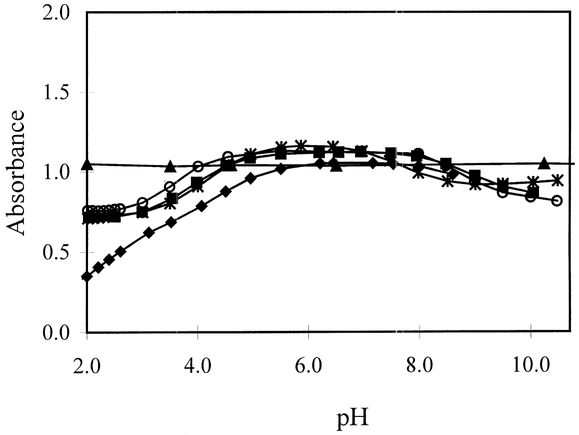


Fig. 3. pH-dependence of absorbance values measured at $\lambda_{max}=420$ nm in iron(III)-DFB (▲), iron(III)-Dha1 (*), iron(III)-Dha2 (○) iron(III)-Dha3 (■), iron(III)-Aha (♦) systems. ($c_{Fe(III)}=4\times10^{-4}$ mol/dm^3, $c_{DFB}=1.0\times10^{-3}$ mol/dm^3, $c_{Dha1}=c_{Dha2}=c_{Dha3}=1.5\times10^{-3}$ mol/dm^3, $c_{Aha}=3.0\times10^{-3}$ mol/dm^3).

Preference for the trihydroxamate-based DFB does not hold for copper(II), which allows the coordination of at most two hydroxamates [13]. The insolubility of the copper(II)-DhaI complexes most probably indicates polynuclear complex formation.

Magnesium(II) forms somewhat more stable complexes than calcium(II) with these ligands. However, hydrolysis, which is significantly more favoured for magnesium(II) than for calcium(II) may compete with complex formation at high pH in the magnesium(II)-containing systems. If this is taken into account, the unexpectedly low stability constant of [MgDFB] (see Table 3) may be explained if it is assumed that the stoichiometry of this species is really [Mg(HDFB)(OH)].

One interesting result is that the formation of calcium(II)-DFB complexes is less favoured than the formation of dihydroxamate complexes, especially with Dha2. The most reasonable explanation of this finding is that the length of the connecting chains allows only the monochelated coordination of DFB to the large calcium(II) ion. The

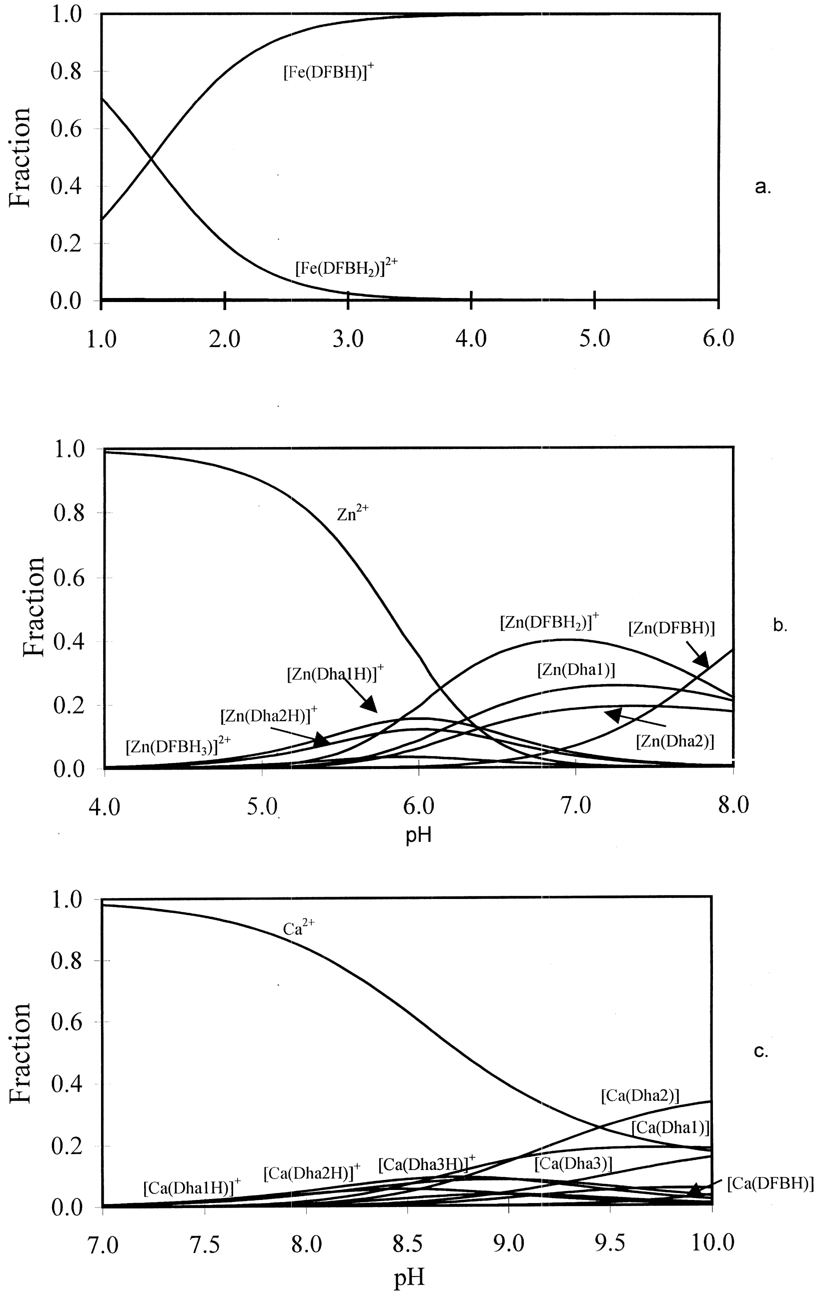


Fig. 4. Concentration distribution curves for hypothtetical systems involving Dha1, Dha2, Dha3, DFB and iron(III) (a), zinc(II) (b) or calcium(II) (c). ($c_{\text{metal}}=2\times10^{-3}$ mol/dm^3, $c_{\text{Dha1}}=c_{\text{Dha2}}=c_{\text{Dha3}}=3\times10^{-3}$ mol/dm^3, $c_{\text{DFB}}=2\times10^{-3}$ mol/dm^3).

amounts of complexes formed in the magnesium(II)-Dha2 and -DFB systems are more comparable, which suggests the formation of a bis-chelated species (at least partly) in [MgHDFB].

It may be seen from the above results that the superposition of different effects (the number of chelating moieties in the ligand, the length and structure of the connecting chain and the ionic radius and oxidation state of the metal) is reflected in the stabilities of the complexes. In order to clarify some effects of the ionic radius of the metal on the basis of our results concentration distributions were calculated for various hypothetical systems. Each hypothetical system was created with involving one of the metal ions and all the studied dihydroxamate-based ligands as well as DFB. The concentrations of the ligands were chosen so that each of them could just complete the octahedral coordination sphere of the metal ion. Of the results, those relating to the iron(III)-, zinc(II)- and calcium(II)-containing systems are shown in Fig. 4. Analysis of the concentration distribution curves in Fig. 4 allows conclusions concerning the preference sequences. In spite of the fact that the stability constants for the complexes formed with Dha1, Dha2 and Dha3 are also involved into the models the iron(III) is coordinated solely by DFB. The larger zinc(II) is distributed between the different ligands (DFB, Dha1 and Dha2) and the calcium(II) (ionic radius 100 pm) forms complexes primarily with Dha2, which has the longest connecting chain.

4. Conclusion

Dha1 and Dha2 were found to be good metal ion sequestering agents. Dha2, having longer connecting chain and higher basicity of the donor atoms forms generally more stable complexes than Dha1. Both are better chelators than the Aha. Dha2 is as good ligand as DFB if the metal ion is copper(II) which allows the coordination of at most two hydroxamates. Dha1 with shorter connecting chain forms insoluble complex(es) with the copper(II). The iron(III) prefers DFB much more, but complexation of the large calcium(II) by Dha1 and especially by Dha2 is significantly more favoured than by DFB.

Acknowledgements

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References

[1] J.B. Neilands, Microbal Iron Metabolism, Academic Press, New York, 1974.

[2] A.L. Crumbliss, in: G. Winkelman (Ed.), CRC Handbook of Microbal Iron Chelates, CRC, New York, 1991.

[3] C.J. Cassano, K.N. Raymond, J. Am. Chem. Soc. 100 (1978) 5371.

[4] S.J. Barclay, P.E. Riley, K.N. Raymond, J. Am. Chem. Soc. 104 (1982) 6802.

[5] Z. Hou, C.J. Sunderland, T. Nishio, K.N. Raymond, J. Am. Chem. Soc. 118 (1996) 5148.

[6] S.J. Barclay, B.H. Huynh, K.N. Raymond, Inorg. Chem. 23 (1984) 2011.

[7] M.A. Santos, M.A. Esteves, M. Candida, T. Vaz, J. Chem. Soc., Dalton Trans. (1993) 927.

[8] A. Evers, R.D. Hancock, A.E. Martell, R.J. Motekaitis, Inorg. Chem. 28 (1989) 2189.

[9] M.T. Caudle, R.D. Stevens, A.L. Crumbliss, Inorg. Chem. 33 (1994) 6111.

[10] M.T. Caudle, L.P. Cogswell III, A.L. Crumbliss, Inorg. Chem. 33 (1994) 4759.

[11] E. Farkas, K. Megyeri, L. Somsák, L. Kovács, J. Inorg. Biochem. 70 (1998) 41.

[12] E. Farkas, E. Kozma, M. Pethö, K.M. Herlihy, G. Micera, Polyhedron 17 (1998) 3331.

[13] E. Farkas, H. Csóka, G. Micera, A. Dessi, J. Inorg. Biochem. (1997) 281.

[14] B. Kurzak, H. Kozlowski, E. Farkas, Coord. Chem. Rev. 114 (1992) 169.

[15] A.H. Blatt, Organic Syntheses, Wiley, New York, 1963.

[16] G. Gran, Acta Chem. Scand. 4 (1950) 599.

[17] H. Irving, M.G. Miles, L.D. Pettit, Anal. Chim. Acta 38 (1967) 475.

[18] L. Zékány, I. Nagypál, in: D. Legett (Ed.), Computational Methods for the Determination of Stability Constants, Plenum Press, New York, 1985.

[19] G.H. Khoe, P.L. Brown, R.N. Sylva, J. Chem. Soc., Dalton Trans. (1986) 1901.