Lariat Ether Carboxylic Acids as Ionizable Hosts in the Second Coordination Sphere of the Siderophore Ferrioxamine B in Chloroform

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A new series of lariat ether carboxylic acids (L_nCOOH) was synthesized with different lengths of side arm (6, 9, and 12 atom spacers between the benzo-18-crown-6 crown ether ring and the carboxylic acid functional group). These lariat ethers were used as neutral and anionic hosts for the molecular recognition of the cationic siderophore, ferrioxamine B. Lariat ether pK_a values were determined by potentiometric titration in 50% methanol (v/v) to be in the range 5.23−5.32. Molecular recognition of ferrioxamine B occurs through second-sphere complexation of the pendant protonated amine ($-(CH_2)_5NH_3^+$) by the lariat ether cavity, utilizing an ion−dipole host−guest interaction to form a supramolecular assembly in wet chloroform. At conditions where the pendant carboxylic acid side arm is not ionized (pH = 3.2), the lariat ethers behave as the parent unsubstituted crown ether structure, benzo-18-crown-6 (B18C6). At pH conditions well above their pK_a values, the lariat ether carboxylic acids function both as a host and as an internal counterion. The stability of this binary assembly, {FeHDFB$^+$,L_nCOO$^-$}, is significantly increased (log K_{app} = 4.85 for $L_{12}COO^-$) compared with that of the ternary assembly involving the protonated lariat ether, {FeHDFB$^+$,L_nCOOH,ClO_4^-} (log K = 3.26 for L_{12}COOH), or the parent crown ether, {FeHDFB$^+$,B18C6,ClO_4^-} (log K = 3.21). Stability was also observed to increase with the length of the side arm from 6 (log K_{app} = 4.29) to 12 atom spacers (log K_{app} = 4.85). We attribute this effect to the increased conformational freedom of the longer arm, which facilitates interaction between the protonated amine site and the carboxylate moiety.

Introduction

Siderophore-mediated iron acquisition by microbes involves the selective chelation of environmental Fe(III), followed by transport and deposition at the cell surface or cell interior.[1−4] Metal−ligand exchange at the cell surface or penetration of the siderophore complex into the cell interior likely involves molecular recognition by a receptor. The mechanism and proteins involved in cellular siderophore uptake have been reviewed.[5] Membrane-bound receptors are involved in an energy-dependent process where recognition has often been shown to be sensitive to the chirality of the metal center and the ligand.[6] Several natural and synthetic macrocyclic and linear molecules, including crown ethers,[7] can function as host recognition factors[8−10] and have been used as models for cell surface receptors. We are engaged in the development of various models for molecular recognition and liquid membrane transport of siderophore complexes.[11] Recent results illustrate that the siderophore ferrioxamine B can be selectively recognized[12] through second-coordination-sphere complexation of the

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(1) Crumbliss, A. L. In *Handbook of Microbial Iron Chelates;* Winkelmann, G., Ed.; CRC Press: Boca Raton, FL, 1991; p 177.
(2) Raymond, K. N.; Telford, J. R. In *Bioinorganic Chemistry: An Inorganic Perspective of Life*; Kessissoglou, D. P., Ed.; NATO ASI Series C: Mathematical and Physical Sciences, 459; Kluwer Academic Publishers: Dordrecht, The Netherlands, 1995; p 25.
(3) Telford, J. R.; Raymond, K. N. *Vol. 1, Molecular Recognition: Receptors for Cationic Guests;* Gokel, G. W., Vol. Ed.; In *Comprehensive Supramolecular Chemistry;* Lehn, J.-M. Exec. Ed.; Pergamon Press: London, 1996; p 245.
(4) Albrecht-Gary, A.-M.; Crumbliss, A. L. In *Iron Transport and Storage in Microorganisms, Plants and Animals;* Sigel, A., Sigel, H., Eds.; Metal Ions in Biological Systems 35; M. Dekker: New York, 1998; p 239.
(5) (a) Crichton, R. R. *Inorganic Biochemistry of Iron Metabolism,* Ellis Horwood: New York, 1991; Chapter 3. (b) Braun, V.; Hantke, K.; Eick-Helmerich, K.; Koster, W.; Pressler, U.; Sauer, M.; Schaeffer, S.; Schoeffler, H.; Staudenmaier, H.; Zimmermann, L. In *Iron Transport in Microbes, Plants and Animals;* Winkelmann, G., van der Helm, D., Nielands, J. B., Eds.; VCH: Weinheim, 1987; p 35. (c) Braun, V.; Hantke, K. In *CRC Handbook of Microbial Iron Chelates;* CRC Press: Boca Raton, FL, 1991; p 107. (d) Earhart, C. In *Iron Transport in Microbes, Plants and Animals;* Winkelmann, G., van der Helm, D., Nielands, J. B., Eds.; VCH: Weinheim, 1987; p 67. (e) Chenault S. S.; Earhart, C. F. *Mol. Microbiol.* **1991**, *5*, 1405. (f) Hantke, K. *Proteins of Iron Storage and Transport;* Elsevier/North Holland: Amsterdam, 1985; p 231. (g) Braun, V.; Gunter, K.; Hantke, K. *Biol. Met.* **1991**, *4*, 14. (h) Koster, W. *Biol. Met.* **1991**, *4*, 23. (i) Lesuisse, E.; Labbe, P. *J. Gen. Microbiol.* **1989**, *135*, 257. (j) Matzanke, B. F.; Berner, I.; Bill, E.; Trautwein, A. X.; Winkelmann, G. *Biol. Met.* **1991**, *4*, 181. (k) Baeumler A. J.; Hantke, K. *Mol. Microbiol.* **1992**, *6*, 1309. (l) Kachadourian, R.; Dellagi, A.; Laurent, J.; Bricard, L.; Kunesch, G.; Expert, D. *BioMetals* **1996**, *9*, 143. (m) Jiang, X.; Payne, M. A.; Cao, Z.; Foster, S. B.; Feix, J. B.; Newton, S. M. C.; Klebba, P. E. *Science* **1997**, *276*, 1261.
(6) Mueller, G.; Matzanke, B. F.; Raymond, K. N. *J. Bacteriol.* **1984**, *160*, 313.
(7) Fyles, T. M. *Top. Inclusion Sci.* **1991**, *2*, 59.
(8) Hiraoka, M., Ed. *Crown Ethers and Analogous Compounds: Studies in Organic Chemistry;* Elsevier: Amsterdam, 1992; Vol. 45.
(9) Cox, B.; Schneider, H. *Coordination and Transport Properties of Macrocyclic Compounds in Solution;* Elsevier: Amsterdam, 1992.
(10) Inoue Y., Gokel, G. W., Eds. *Cation Binding by Macromolecules;* Marcel Dekker Inc.: New York and Basel, 1990.
(11) Crumbliss, A. L.; Batinić-Haberle, I.; Spasojević, I. *Pure Appl. Chem.* **1996**, *68*, 1225.

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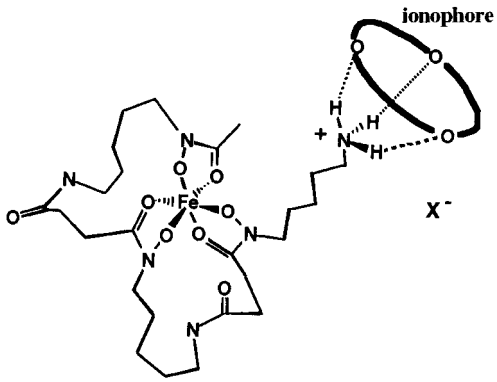
protonated amine side chain by different ionophore host molecules (**I**).[13,14]



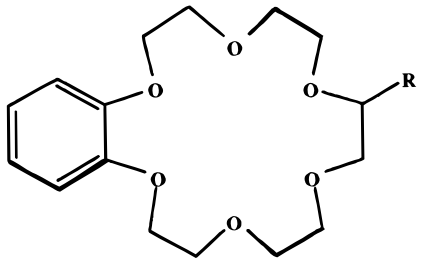
I

Ferrioxamine B second-sphere host−guest complex

Certainly, siderophore cell receptors are more complex than crown ether hosts. However, investigation of synthetic macrocycle hosts will help to elucidate host−guest interactions that are relevant to the recognition and reactivity of stable metal complexes. Such studies are also relevant to how antibiotics and host molecules may influence the biodistribution of Fe carriers such as ferrioxamine B in the body. In an attempt to understand some of the molecular recognition processes that are operative in biological metal-transport systems, we have explored factors that may contribute to the enhanced stability of specific host−guest interactions in the second coordination sphere of a transition metal carrier complex. Structural features associated with the host molecules, as well as some aspects of the metal complex guest, have been explored. The influence of the counterion needed to achieve a neutral supramolecular assembly involving the cationic ferrioxamine B complex has been recognized,[15] as well as the influence of the cation guest.[12,13] The steric requirements of the bulky ferrioxamine B guest and of the host, which influence the stability of the supramolecular assembly, have also been addressed.[16] Since the medium is known to influence the stability of a supramolecular assembly,[17,18] solvent effects have also been investigated.[19] Our previous results demonstrate that, by optimizing the characteristics of all species interacting in the supramolecular assembly (cation guest, macrocyclic host, counterion, and solvent), the stability constant for second-sphere complexation of ferrioxamine B may be enhanced by several orders of magnitude.

A three-dimensional host provides additional opportunities for molecular recognition, facilitated, for example, by additional flexibility, increased number and type of host−guest interaction sites, and desolvation. The cryptands have been compared with crown and aza crown ethers in this regard. The lariat ether concept, with additional donors on a side arm attached to a macrocycle, has been developed by Gokel and others.[20−27] The typical lariat ether arrangement where ring−side arm cooperativity is achieved minimizes the number of kinetic steps necessary for complexation. This leads to an increased complexation rate, which, in combination with a decreased decomplexation rate, leads to a higher cation binding constant. Due to the lower rigidity of the three-dimensional structure formed by a flexible lariat ether side arm, the decomplexation rate is less affected than in the case of cryptands, which makes lariat ethers potentially more suitable as biological carriers. In addition, the possibility of structural variation of the side arm in terms of length and functional group(s) extends the capability for molecular recognition over that of an unsubstituted macrocyclic ionophore.

In the present study, we have designed a series of lariat ethers (**IIb−d**) possessing a functional group (carboxylic acid) capable of ionizing over the pH range investigated. When the lariat



II

IIa, R = H

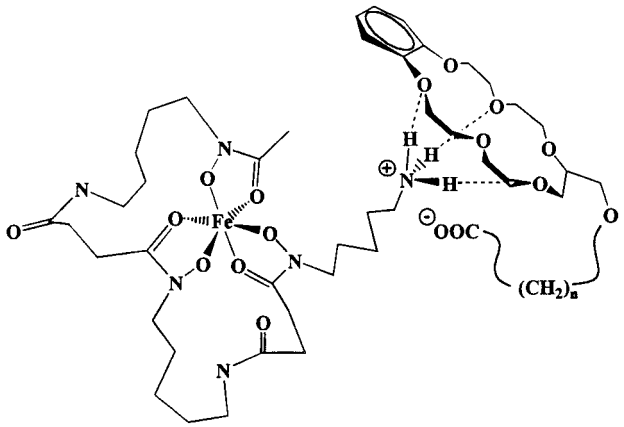
IIb, R = $CH_2O(CH_2)_4COOH$ (L_6COOH)

IIc, R = $CH_2O(CH_2)_7COOH$ (L_9COOH)

IId, R = $CH_2O(CH_2)_{10}COOH$ ($L_{12}COOH$)

ether carboxylic acid is ionized, it can act as a counterion. Consequently, as illustrated in **III**, ion−ion host−guest interactions are added to the existing ion−dipole interactions, maximizing binding dynamics.[28,29] We have developed a synthetic strategy that allows us to tether a specific functional group to a crown ether cavity using different tether lengths. This enables us to probe the influence of tethering the counteranion to the macrocycle host and to determine the optimum tether length

(12) Spasojević, I.; Batinić-Haberle, I.; Choo, P. L.; Crumbliss, A. L. *J. Am. Chem. Soc.* **1994**, *116*, 5714.
(13) Batinić-Haberle, I.; Spasojević, I.; Crumbliss, A. L. *Inorg. Chem.* **1996**, *35*, 2352.
(14) Caldwell, C. D.; Crumbliss, A. L. *Inorg. Chem.*, accepted for publication.
(15) Batinić-Haberle, I.; Crumbliss, A. L. *Inorg. Chem.* **1995**, *34*, 928.
(16) Batinić-Haberle, I.; Spasojević, I.; Bartsch, R. A.; Crumbliss, A. L. *J. Chem. Soc. Dalton Trans.* **1995**, 2503.
(17) Ovchinikov, Yu. A.; Ivanov, V. T.; Shkrob, A. M. *Membrane-Active Complexones;* Elsevier Scientific Publishing: Amsterdam, 1974.
(18) Cox, B.; Schneider, H. *Coordination and Transport Properties of Macrocyclic Compounds in Solution;* Elsevier: Amsterdam, 1992.
(19) Batinić-Haberle, I.; Spasojević, I.; Crumbliss, A. L. *Inorg. Chim. Acta* **1997**, *260*, 35.
(20) Gokel, G. W. In *Lariat Ethers in Inclusion Compounds, Vol. 4, Key Organic Host Systems;* Atwood, J. L., Davies, J. E. D., MacNicol, D. D., Eds.; Oxford University Press: Oxford, 1991; p 283.
(21) Gokel, W.; Nakano, A. In *Crown Compounds. Towards Future Applications;* Cooper, S. R., Ed.; VCH Publishers: New York, 1992; p 1.
(22) Gokel, G. W. *Chem. Soc. Rev.* **1992,** *21*, 39.
(23) Weber, E. *New Development in Crown Ether Chemistry: Lariat, Spherand and Second-Sphere Complexes in Crown Ethers and Analogues;* Patai, S., Rappoport, Z., Eds.; John Wiley & Sons: Chichester, 1989.
(24) Gokel, G. W.; Arnold, K. A.; Delgado, M.; Echeverria, L.; Gatto, V. J.; Gustowski, D. A.; Hernandez J.; Kaifer, A.; Miller, S. R.; Echegoyen, L. *Pure Appl. Chem.* **1988**, *60*, 461.
(25) Li, Y.; Gokel, G.; Hernandez, J.; Echegoyen, L. *J. Am. Chem. Soc.* **1994**, *116*, 3087.
(26) Tsukube, H. *Coord. Chem. Rev.* **1996**, *148*, 1.
(27) Potvin, P. G.; Lehn, J.-M. In *Progress in Macrocyclic Chemistry. Vol. 3;* Izatt, R M., Christensen, J. J., Eds.; Wiley: New York, 1987; p 167.
(28) Koszuk, J. F.; Czech, B. P.; Walkowiak, W.; Babb, D. A.; Bartsch, R. A. *J. Chem. Soc., Chem. Commun.* **1984,** 1504.
(29) Strzelbicki, J.; Charevicz, A.; Bartsch, R. A. *J. Inclusion Phenom.* **1988,** *6*, 57.



III

Supramolecular assembly involving lariat ether carboxylic acid (host) and ferrioxamine B (guest) {FeHDFB$^+$,L$_n$COO$^-$}

for stable host−guest complex formation. We have selected lariat ethers based on the benzo-18-crown-6 crown ether structure, since we have previously found this host cavity to form stable host−guest complexes with ferrioxamine B.[13]

In previous reports from our laboratory,[12−16] we used what may be termed a "neutral ionophore model". This model applies to an aqueous/organic extraction process in which a neutral ionophore binds to a cation that has been distributed from the aqueous phase together with its accompanying anion. The overall extraction equilibrium (eq 1)

$$\mathrm{FeHDFB^+_{aq} + anion^-_{aq} + ionophore^0_{org} \rightleftarrows \{FeHDFB^+,ionophore^0,anion^-\}_{org} \quad K_{ex} \quad (1)}$$

may be broken down into a cation−anion pair distribution (eq 2)

$$\mathrm{FeHDFB^+_{aq} + anion^-_{aq} \rightleftarrows \{FeHDFB^+,anion^-\}_{org} \quad K_d \quad (2)}$$

and subsequent association of the ionophore with the ion pair in the organic phase (eq 3).

$$\mathrm{\{FeHDFB^+,anion^-\}_{org} + ionophore^0_{org} \rightleftarrows \{FeHDFB^+,ionophore^-,anion^-\}_{org} \quad K \quad (3)}$$

To express the stability of the lariat ether binary assembly {FeHDFB$^+$,L$_n$COO$^-$} in the form of an association constant as in the neutral ionophore model, the distribution of ferrioxamine B is expressed here as a distribution ratio, *D*, and not as a distribution constant, K_d (vide infra). As a result, the association constant for the {FeHDFB$^+$,L$_n$COO$^-$} assembly is an *apparent* constant, K_{app}, that is valid at the constant perchlorate concentration (0.08 M) used in our experiments. This may be described as an "anionic ionophore model". Since the ionophore is negatively charged, a model for the extraction process does not need to involve the accompanying anion. The overall extraction equilibrium may be written as eq 4.

$$\mathrm{FeHDFB^+_{aq} + ionophore^-_{org} \rightleftarrows \{FeHDFB^+,ionophore^-\}_{org} \quad K_{ex} \quad (4)}$$

which may be divided into the cation distribution into the organic phase (eq 5)

$$\mathrm{FeHDFB^+_{aq} \rightleftarrows FeHDFB^+_{org} \quad D \quad (5)}$$

and the host−guest association of cation and anionic ionophore to form a supramolecular assembly (eq 6).

$$\mathrm{FeHDFB^+_{org} + ionophore^-_{org} \rightleftarrows \{FeHDFB^+,ionophore^-\}_{org} \quad K_{app} \quad (6)}$$

The first two equilibria in each model can be quantified experimentally, and the host−guest association constant (a quantity by which the neutral and charged ionophores are readily compared) can be calculated as $K = K_{ex}/K_d$ (eq 3) for the neutral ionophore model and $K_{app} = K_{ex}/D$ (eq 6) for the anionic ionophore model.

The data reported here, to the best of our knowledge, provide the first example of the formation of a stable supramolecular assembly involving second-coordination-sphere complexation of a cationic transition metal complex with a lariat ether which functions as both a host and a counterion.

Experimental Section

Materials. Benzo-18-crown-6 (B18C6, **IIa**) was used as obtained from Aldrich (ϵ = 2200 M^{-1} cm^{-1} at 278 nm[13]). Ethyl ω-bromoalkanoates were prepared from commercially available ω-bromoalkanoic acids (Aldrich) by esterification with *p*-toluenesulfonic acid catalyst in EtOH−benzene (1:1).[30] Lariat ether carboxylic acids (L$_n$COOH), a new series of B18C6 carboxylic acids, with 6 (**IIb**, L$_6$COOH), 9 (**IIc**, L$_9$COOH), and 12 (**IId**, L$_{12}$COOH) atom spacers between the crown ether ring and the carboxylic acid group were synthesized as described below. The concentrations of crown and lariat ethers were varied between 5.0 × 10^{-3} and 5.0 × 10^{-2} M, depending on their distribution patterns and their ability to extract ferrioxamine B from the aqueous to the chloroform phase. Mg(ClO$_4$)$_2$ (99%) and Mg(OH)$_2$ (98%) were purchased from Aldrich and used for maintaining the ionic strength at 0.1 M and the desired pH. Ferrioxamine B perchlorate solutions were prepared as described previously.[15] ***Warning:*** *Extreme care should be taken when working with perchlorate salts!* The concentrations of FeHDFB$^+$, ClO$_4^-$, and Mg^{2+} in the aqueous solutions were 2.0 × 10^{-2}, 8.0 × 10^{-2}, and 3.0 × 10^{-2} M, respectively. Twice-distilled water and chloroform from Fisher Scientific (spectroscopic grade) were used throughout the experiments. Physical measurements were made using aqueous solutions saturated with chloroform and chloroform solutions saturated with water. Infrared spectra were measured with a Perkin-Elmer 1600 infrared spectrophotometer. ^{1}H NMR spectra were taken with an IBM AF-200 spectrometer and are given in ppm (δ) downfield from TMS. UV/visible spectra were measured with a HP 8451A diode array spectrophotometer. Elemental analyses were performed by Desert Analytics Laboratory, Tucson, AZ.

L$_6$COOH (IIb), 5-[11′-(Oxymethyl)-2′,3′-benzo-18-crown-6]pentanoic Acid. NaH (0.71 g, 60% in mineral oil, 17.8 mmol) was washed with dry pentane under nitrogen, and a solution of 1.22 g (3.56 mmol) of 11-(hydroxymethyl)-2,3-benzo-18-crown-6[31] (**II**, with R = CH$_2$OH) in 30 mL of THF was added. The mixture was stirred for 2 h at room temperature, and a solution of 1.12 g (0.83 mmol) of ethyl 5-bromopentanoate in 50 mL of THF was added during a 2-h period, followed by refluxing overnight. After the mixture was cooled to 0 °C in an ice bath, water was carefully added to destroy the excess NaH. The THF was evaporated in vacuo, and 300 mL of water was added to the residue. The aqueous mixture was extracted with EtOAc (3 × 100 mL). The aqueous layer was acidified to pH 1 with 6 M HCl and extracted with CH$_2$Cl$_2$ (3 × 100 mL). The combined organic extracts (EtOAc and CH$_2$Cl$_2$) were dried over MgSO$_4$ and evaporated in vacuo.

(30) Leonard, N. J.; Fox, R. C.; Oki, M. *J. Am. Chem. Soc.* **1954,** *76*, 5708.
(31) Czech, B.; Babb, D. A.; Bartsch, R. A. *Org. Prep. Proced. Int.* **1983**, *15*, 29.

The yellowish oil was purified by evaporation to remove impurities from the product in a simple distillation apparatus by heating at 80−85 °C/0.5 Torr for 1 day to provide 0.94 g (60%) of **IIb** as a pale yellow oil.[32] ^{1}H NMR ($CDCl_3$): δ 1.63−1.66 (m, 4H), 2.28−2.38 (t, 3H), 3.45−3.94 (m, 23H), 4.13−4.17 (m, 4H), 4.50−5.40 (br s, 1H), 6.89 (s, 4H). IR (deposited on a NaCl plate from CH_2Cl_2 solution): 3437 (O−H), 1729 (C═O), 1256, 1124 (C−O) cm^{-1}. UV/vis: molar absorptivity 2300 M^{-1} at 278 nm. Anal. Calcd for $C_{22}H_{34}O_9$: C, 59.71; H, 7,74. Found: C, 59.54; H, 7.46.

L_9COOH (IIc), 8-[11′-(Oxymethyl)-2′,3′-benzo-18-crown-6]octanoic Acid. IIc was prepared by the same procedure as given above for the synthesis of **IIb**, with the exceptions that ethyl 8-bromooctanoic acid was used as the alkylating agent and the crude product was purified by evaporation of impurities at 120−125 °C/0.5 Torr for 2 days. A 46% yield of **IIc** was isolated as a yellow oil. ^{1}H NMR ($CDCl_3$): δ 1.31−1.53 (m, 10H), 2.29−2.37 (t, 2H), 3.37−3.46 (m, 5H), 3.61−3.94 (m, 14H), 4.13−4.17 (m, 4H), 6.89 (s, 4H). IR (deposited on a NaCl plate from CH_2Cl_2 solution): 3367 (O−H), 1733 (C═O), 1256, 1124 (C−O) cm^{-1}. UV/vis: molar absorptivity 2200 M^{-1} at 278 nm. Anal. Calcd for $C_{25}H_{40}O_9$: C, 61.97; H, 8.32. Found; C, 62.37; H, 8.21.

L_{12}COOH (IId), 11-[11′-(Oxymethyl)-2′,3′-benzo-18-crown-6]-undecanoic acid. IId was prepared by the same procedure as given above for the synthesis of **IIb**, with the exceptions that ethyl 11-bromoundecanoic acid was used as the alkylating agent and the crude product was purified by evaporation of impurities at 130−135 °C/0.5 Torr for 1 day. A 47% yield was obtained as a pale brown oil. ^{1}H NMR ($CDCl_3$): δ 1.27−1.62 (m, 16H), 2.29−2.37 (t, 3H), 3.38−3.47 (m, 4H), 3.67−3.94 (m, 15H), 4.13−4.17 (m, 4H), 6.90 (s, 4H). IR (deposit from CH_2Cl_2 solution on a NaCl plate): 3508 (O−H), 1730 and 1708 (C═O), 1256 and 1125 (C−O) cm^{-1}. UV/vis: molar absorptivity 2700 M^{-1} at 278 nm. Anal. Calcd for $C_{28}H_{46}O_9$: C, 63.86; H, 8.80. Found: C, 63.47; H, 8.61.

Methods. Two-phase distribution and extraction experiments were typically performed by vigorously agitating equal volumes of aqueous (with $FeHDFB^+$) and chloroform (with or without ionophore) solutions in polyethylene-capped glass vials. Short-time centrifugation (minutes) was followed by long-time equilibration (hours), after which the phases were carefully separated. In many instances, spectrophotometric measurements of the chloroform phase were avoided by using a re-extraction method in which the separated chloroform phase was put in contact with "fresh" aqueous solution, typically containing 0.1 M KNO_3. Due to the high affinity of K^+ for 18-atom cavity size crown ethers, all of the ferrioxamine B is displaced by K^+. As a consequence, the ferrioxamine B is quantitatively re-extracted by the aqueous phase due to a very low distribution constant, K_d (vide infra, Table 2). Additional experimental details and modifications concerning a particular system are given in the Results section.

Potentiometric titration of the lariat ether carboxylic acids in 50% MeOH/aqueous (v/v) solution was performed in the pH region from 2 to 12 under a purified N_2 atmosphere in a thermostated cell at 25 ± 0.1 °C. The solution was titrated with 0.1 M NaOH, prepared by dilution of a 0.2 M NaOH standardized solution with MeOH in 1:1 ratio by volume. The pH measurements were carried out using a Corning pH-meter with a Corning combined glass electrode containing 0.1 M NaCl in the reference electrode compartment. Internal calibration of the electrode (using data from a separate experiment in which perchloric acid in 50% methanol was titrated with NaOH in 50% methanol) and pK_a value calculation (in $[H^+][L^-]/[HL]$ mode) were performed through SUPERQUAD-MAGEC[33] cycling refinement.[34]

Results

Lariat Ethers. Compounds in the series of benzo-18-crown-6-carboxylic acids (**IIb**−**d**) with varying chain lengths between the crown ether ring and the carboxylic acid function were prepared in 60, 46, and 47% yields, respectively, as shown in Scheme 1. An excess of sodium hydride in the reaction mixture promoted hydrolysis of the first-formed lariat ether esters to give the lariat ether carboxylic acids directly.

Scheme 1

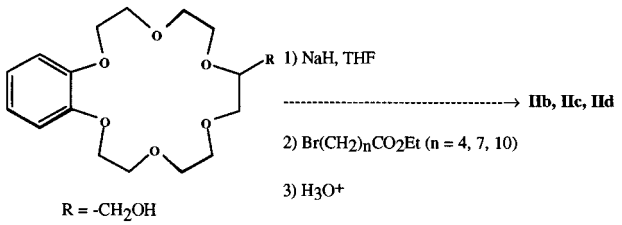


Table 1. Acid Dissociation Constants for Lariat Ether Carboxylic Acids in 50% Methanol at 25 ± 0.1 °C[a]

lariat ether carboxylic acid	pK_a[b]
IIb, L_6COOH	5.23 ± 0.02
IIc, L_9COOH	5.27 ± 0.04
IId, L_{12}COOH	5.32 ± 0.04

[a] I = 0.1 (HCl/NaCl). [b] Concentration constants; $[H^+][L^-]/[HL]$ mode.

Acid dissociation constants for the carboxylic acid side chains of the lariat ethers (**IIb**−**d**) were determined by potentiometric titrations in 50% methanol (by volume). The pK_a values obtained are in the expected range[35] for a carboxylic acid in 50% methanol (Table 1). Variations in lariat ether chain length do not significantly influence the pK_a values.

Distribution, Extraction, and Host−Guest Equilibria. The overall extraction of ferrioxamine B by the ionophore from the aqueous phase to the chloroform phase, when corrected for the independent distribution of ferrioxamine B and the ionophore between the two phases, enables us to calculate a "host−guest" association constant for supramolecular assembly formation in the chloroform phase. Our extraction studies were conducted at low (3.2) and high (9.3) pH using the buffer system described below. At low pH, the lariat ether carboxylate is undissociated and behaves as an unsubstituted crown ether that requires a counterion (ClO_4^-) for charge neutrality. Treatment of the host−guest association equilibrium in this case follows the neutral ionophore model described in eqs 1−3. At high pH, the carboxylate group is dissociated, and the lariat ether acts as an ionophore cavity and a counterion for the positively charged $FeHDFB^+$. Treatment of the host−guest association equilibrium in this case follows the anionic ionophore model described in eqs 4−6.

(i) Buffer System. A major challenge of the phase equilibria experiments was to maintain a constant pH in an appropriate range that would ensure the deprotonation of the lariat ether carboxylic acid group (pK_a = 5.23−5.32), yet maintain protonation at the pendant amine site of ferrioxamine B (pK_a = 10.40).[36] $Mg(OH)_2$ proved to be an excellent choice as a pH = 9.3 "buffer" for the aqueous phase due to its limited solubility[37] and its minimal distribution, which results in negligible extraction into chloroform.[15] NaOH was used to control pH in experiments carried out at pH = 11.3 in order to study the deprotonated form of ferrioxamine B ($FeDFB^0$).

(32) Under these conditions, the ethyl ester which was formed by the alkylation reaction was automatically hydrolyzed to the desired carboxylic acid.

(33) Leggett, D. J., Ed. *Computational Methods for the Determination of Formation Constants;* Plenum Press: New York, 1985; p 37.

(34) May, P. M.; Williams, D. R. *Talanta* **1982**, *29*, 249.

(35) Rorabacher, D. B.; MacKellar, W. J.; Shu, F. R.; Sister Bonavita, M. *Anal. Chem.* **1971**, *43*, 561.

(36) Evers, A.; Hancock, R. D.; Martell, A. E.; Motekaitis, R. J. *Inorg. Chem.* **1989**, *28*, 2189.

(37) *Stability Constants of Metal Complexes. Part A: Inorganic Ligands;* Hogfeldt, E., Compiler; IUPAC Chemical Data Series 21; Pergamon Press: Oxford, 1982.

Table 2. Distribution Equilibrium Constants between Water and Chloroform at 25 ± 0.5 °C for Ferrioxamine B, Crown Ether, and Lariat Ether Carboxylic Acids[a]

	Ferrioxamine B	
	K_d	D
$FeHDFB^+$	3.1×10^{-4} [b]	2.5×10^{-5} [c]
$FeDFB^0$	1.6×10^{-2} [d]	
	Lariat and Crown Ethers	
	K_d',[e] pH = 3.2	D' [f] pH = 9.3
IIa, B18C6	3.03×10^{-3} [g]	
IIb, L_6COOH	5.48×10^{-3}	2.13
IIc, L_9COOH	1.04×10^{-3}	0.49
IId, $L_{12}COOH$	1.09×10^{-3}	0.09

[a] $I = 0.1$ ($FeHDFB^+$,Mg^{2+},ClO_4^-). The values reported are based on 3−5 independent experiments, and the errors are <5% for lariat ether and <10% for $FeHDFB^0$ distribution. [b] K_d, M^{-1}; eq 7, data from ref 12. [c] Calcd by eq 8 as $K_d[ClO_4^-]_{aq}$, where $[ClO_4^-]_{aq} = 0.08$ M. [d] Defined in eq 9. [e] Defined in eq 10. [f] Defined in eq 11. [g] Data from ref 13.

(ii) Distribution Equilibria. (a) Ferrioxamine B ($FeHDFB^+$). At aqueous pH = 3.2, the distribution of $FeHDFB^+$ as an ion pair {$FeHDFB^+$,ClO_4^-} into chloroform may be expressed as in eq 7. This value has been determined previously (Table 2).[12]

$$K_d = [\{FeHDFB^+,ClO_4^-\}]_{org}/([FeHDFB^+]_{aq}[ClO_4^-]_{aq}) \quad (7)$$

At aqueous pH = 9.3, for the anionic ionophore model, K_d values were used to calculate a distribution ratio, D (eq 8), by

$$D = [FeHDFB^+]_{org}/[FeHDFB^+]_{aq} = K_d[ClO_4^-] \quad (8)$$

multiplying the K_d value by the perchlorate concentration, $[ClO_4^-] = 0.08$ M, that was used in the distribution and extraction experiments. The symbol D was used to distinguish the distribution ratio from the distribution constant K_d, which is a true equilibrium constant. The value calculated for D is given in Table 2.

(b) Deprotonated (Uncharged) Ferrioxamine B ($FeHDFB^0$). We have performed experiments to address the distribution characteristics of the deprotonated form of ferrioxamine B, $FeDFB^0$, and its ability to form a host−guest complex with L_nCOOH. This is important because, at the conditions used to investigate the host characteristics of the proton-dissociated lariat ethers (pH = 9.3/$Mg(OH)_2$ buffer), some ferrioxamine B is present in its deprotonated form, $FeDFB^0$ ($pK_a = 10.40$).[36] The lack of positive charge and the increase in hydrophobicity are expected to affect the distribution and, consequently, the extraction equilibria of ferrioxamine B. The distribution (eq 9) was determined using a ferrioxamine B

$$K_d = [FeDFB^0]_{org}/[FeDFB^0]_{aq} \quad (9)$$

solution brought to pH = 9.3 by the addition of $Mg(OH)_2$. At that pH, the fraction of $FeDFB^0$ in the total ferrioxamine B concentration was determined using $pK_a = 10.40$. The experiment was performed in such a way that equal volumes of the pH = 9.3 aqueous solution and chloroform were agitated vigorously for 10 min and left to equilibrate for 14 h. After equilibration, an aliquot of the chloroform layer was mixed with an aqueous solution containing 0.1 M KNO_3, whereupon ferrioxamine B ($FeHDFB^+$ and $FeDFB^0$) was re-extracted back into the aqueous phase, where its concentration was determined spectrophotometrically. The data were corrected for the distribution of $FeHDFB^+$. An independent experiment was used to establish that the molar absorptivity of ferrioxamine B ($\epsilon_{425} = 2600$ M^{-1} cm^{-1})[38] does not change upon deprotonation of the amine site. The distribution data given in Table 2 show that the $FeDFB^0$ distributes into chloroform much more readily than $FeHDFB^+$.

(c) Lariat Ether Carboxylic Acids (L_nCOOH). At pH = 3.2, the lariat ethers were not deprotonated, and, therefore, their distribution was determined as previously described for B18C6.[13] An aqueous solution (pH = 3.2/$HClO_4$) was equilibrated with a chloroform solution of lariat ether carboxylic acid. After equilibration, the aqueous lariat ether was re-extracted back into chloroform, where its concentration was determined spectrophotometrically. The distribution equilibrium constant, K_d', as defined in eq 10, was determined over a wide range of total lariat ether concentrations between 5×10^{-3} and 5×10^{-2} M and is given in Table 2.

$$K_d' = [L_nCOOH]_{aq}/[L_nCOOH]_{org} \quad (10)$$

At pH = 9.3, the lariat ether carboxylic acid functional group is fully deprotonated (Table 1). Deprotonation of the lariat ether followed by its distribution, defined in eq 11, was accomplished

$$D' = [L_nCOO^-]_{aq}/[L_nCOO^-]_{org} \quad (11)$$

by vigorously shaking equal volumes of aqueous and chloroform layers (usually 2 mL) for 10 min after ca. 15 mg of $Mg(OH)_2$ had been added to maintain the aqueous phase pH at 9.3. The concentration of the lariat ether carboxylic acids in the organic phase was determined spectrophotometrically, while their aqueous phase concentrations were calculated as the difference between the total and chloroform phase concentrations. D' values, determined in the range of lariat ether concentrations between 5×10^{-3} and 5×10^{-2} M, are given in Table 2. These D' values are used to calculate the total organic phase equilibrium lariat ether carboxylic acid concentrations and, consequently, the uncomplexed ionophore in the extraction equilibrium.

A separate experiment was carried out to confirm that the high affinity of the hydrophilic $FeHDFB^+$ for the aqueous phase does not affect the distribution of the lariat ether between the chloroform and aqueous phases through an interaction with $FeHDFB^+_{aq}$. After the extraction equilibrium of $FeHDFB^+$ was achieved as described below, the lariat ether carboxylic acid concentration in the organic phase was determined and its aqueous phase concentration calculated. The D' values for the lariat ethers obtained in the extraction experiment are equal (within 1%) to the D' values obtained in the absence of ferrioxamine B in the distribution experiment. Consequently, the association of $FeHDFB^+$ with lariat ether carboxylic acid in the aqueous phase does not occur on an observable level. This is in agreement with literature data that report several orders of magnitude difference between the association of certain cations with crown ethers in apolar and polar media.[39,40]

(iii) Extraction Equilibria. The extraction of ferrioxamine B by the lariat ethers at aqueous pH = 3.2 and 9.3 ($Mg(OH)_2$ buffer) was performed using the same procedure as previously described for crown ethers.[12,13] When $Mg(ClO_4)_2$ and $NaClO_4$

(38) Monzyk, B.; Crumbliss, A. L. *J. Am. Chem. Soc.* **1982**, *104*, 4921.
(39) Izatt, R M.; Bradshaw, J. S.; Nielsen, S. A.; Lamb, J. D.; Christensen, J. J. *Chem. Rev.* **1985**, *85*, 271.
(40) Izatt, R. M.; Pawlak, K.; Bradshaw, J. S.; Bruening, R. L. *Chem. Rev.* **1991**, *91*, 1721.

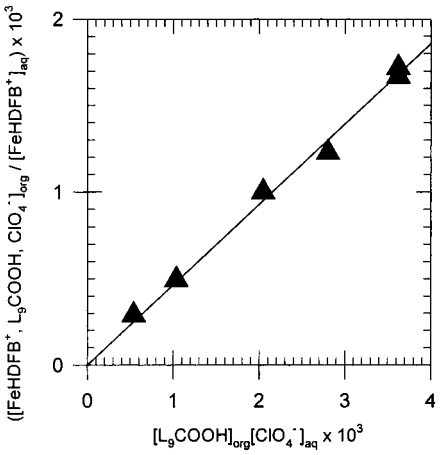


Figure 1. Plot of $[\{FeHDFB^+,L_9COOH,ClO_4^-\}]_{org}/[FeHDB^+]_{aq}$ as a function of $(L_9COOH]_{org}[ClO_4^-]_{aq})$ according to eq 13 for the $FeHDFB^+$ extraction with ClO_4^- by L_9COOH into chloroform at aqueous pH = 3.2, $I = 0.1$ ($FeHDFB^+$,Mg^{2+},ClO_4^-), $T = 25 \pm 0.1$ °C.

are used as supporting electrolyte, their extraction equilibria relative to that of ferrioxamine B may be neglected.[15,41,42]

At aqueous pH = 3.2, the neutral ionophore model applies (eqs 1−3), and the specific overall extraction equilibrium investigated here may be represented as shown in eqs 12 and 13. The equilibrium concentrations were calculated as follows.

$$FeHDFB^+_{aq} + ClO_4^-_{aq} + L_nCOOH_{org} \overset{K_{ex}}{\rightleftharpoons} \{FeHDFB^+,L_nCOOH,ClO_4^-\}_{org} \quad (12)$$

$$K_{ex} = [\{FeHDFB^+,L_nCOOH,ClO_4^-\}]_{org}/([FeHDB^+]_{aq}[L_nCOOH]_{org}[ClO_4^-]_{aq}) \quad (13)$$

$[FeHDFB^+]_{aq}$ and $[ClO_4^-]_{aq}$ were calculated as the difference between their total and organic phase concentrations. The organic phase $FeHDFB^+$ concentration was measured spectrophotometrically ($\epsilon = 2600\ M^{-1}\ cm^{-1}$)[38] after being re-extracted into the aqueous phase by 0.1 M KNO_3. The $[L_nCOOH]_{org}$ was calculated as the difference between its total concentration and its concentration in the $\{FeHDFB^+,L_nCOOH,ClO_4^-\}_{org}$ assembly, corrected by its distribution constant, K_d'.

The lariat ethers **IIb**−**d** were found to be effective hosts for the extraction of aqueous ferrioxamine B into chloroform. A plot of $[\{FeHDFB^+,L_nCOOH,ClO_4^-\}]_{org}/\{[FeHDFB^+]_{aq}$ vs $([L_nCOOH]_{org}[ClO_4^-]_{aq})$ according to eq 13 at pH = 3.2 yields a straight line for all three of the lariat ethers investigated. A representative plot for lariat ether **IIc** is shown in Figure 1. This establishes the validity of the 1:1 host−guest stoichiometry $\{FeHDFB^+,L_nCOOH,ClO_4^-\}$ shown in eq 12 and permits calculation of the extraction equilibrium constant (K_{ex}) from the slope of the plot (Table 3).

At aqueous pH = 9.3, the anionic ionophore model applies (eq 4−6), and the specific extraction equilibrium may be represented by eqs 14 and 15. The extraction experiment was performed in the same way as the distribution of the lariat ether carboxylic acid at pH = 9.3. The $[FeHDFB^+]_{aq}$ was calculated as the difference between its total and organic phase concentrations. The $[L_nCOO^-]_{org}$ concentration was calculated as the difference between its total concentration in the organic phase (as given by D') and its concentration in the form of the host−guest assembly, $\{FeHDFB^+,L_nCOO^-\}_{org}$. The data have been corrected for the presence of $[FeDFB^0]_{org}$ that does not associate with the lariat ethers but is present in the organic phase due to its relatively high distribution (see Table 2).

Table 3. Extraction Equilibrium Constants (log K_{ex}, Eqs 13 and 15) for Ferrioxamine B with Crown Ether and Lariat Ether Carboxylic Acid Hosts in Chloroform at 25 ± 0.1 °C[a]

	log K_{ex}	
host	pH = 3.2	pH = 9.3
IIa, B18C6	−0.30[b]	−0.34
IIb, L_6COOH	−0.13	−0.30
IIc, L_9COOH	−0.33	−0.05
IId, $L_{12}COOH$	−0.25	0.26

[a] $I = 0.1$ ($FeHDFB^+$,Mg^{2+},ClO_4^-). Values reported are based on 3−5 independent determinations, and the errors are <10%. [b] Data from ref 13.

$$FeHDFB^+_{aq} + L_nCOO^-_{org} \overset{K_{ex}}{\rightleftharpoons} \{FeHDFB^+,L_nCOO^-\}_{org} \quad (14)$$

$$K_{ex} = [\{FeHDFB^+,L_nCOO^-\}]_{org}/([FeHDFB^+]_{aq}[L_nCOO^-]_{org}) \quad (15)$$

The lack of $FeDFB^0$ association with the lariat ethers was verified in a separate extraction experiment at pH > 11 (maintained by NaOH), where >80% of ferrioxamine B exists as the deprotonated form, $FeDFB^0$.[36] Changing the supporting electrolyte cation from Mg^{2+} to Na^+ was not expected to influence the association of ferrioxamine B with the lariat or crown ether, as their perchlorate salts do not distribute between water and chloroform on an observable level[15] and, thus, are not extracted into chloroform. (To confirm this assertion, we established that the host−guest association constant for $FeHDFB^+$ complexation by B18C6 was the same when either $Mg(ClO_4)_2$ or $NaClO_4$ was used as a supporting electrolyte at pH = 3.2.) When ferrioxamine B is uncharged ($FeDFB^0$), the lariat ether side arm is not needed as a counterion. Consequently, B18C6 (**IIa**) may be used instead of the lariat ether carboxylic acid in order to simplify the experiment. There was no observable difference in the ferrioxamine B concentration in the chloroform phase with and without B18C6, and, consequently, there is no evidence for host−guest complex formation between $FeDFB^0$ and B18C6. Thus, we may safely assume that $FeDFB^0$ does not associate with lariat ethers. These observations are consistent with a report that no association of the substituted neutral amine $PhCH_2NH_2$ was observed with oxo-18C5, although a strong interaction was observed for $R_2NH_2^+$.[43] Since the numbers of hydrogen atoms available for hydrogen-bonding interactions with the crown ether ring are equal in both cases, it appears that positive charge is a prerequisite for a substituted amine cation's ability to interact with crown ethers.

At pH = 9.3, the lariat ethers are deprotonated and can serve as both host and counteranion. A plot of $[\{FeHDFB^+,L_n$-

(41) Lamb, J. D.; Christensen, J. J.; Izatt, S. R.; Bedke, K.; Astin, M. S.; Izatt, R. M. *J. Am. Chem. Soc.* **198**0, *102*, 3399.
(42) Christensen, J. J.; Lamb, J. D.; Izatt, S. R.; Starr, S. E.; Weed, G. C.; Astin, M. S.; Stitt, B. D.; Izatt, R. M. *J. Am. Chem. Soc.* **1978**, *100*, 3219.
(43) Beresdorf, G. D.; Stoddart, J. F. *Tetrahedron Lett.* **198**0, *21*, 867.

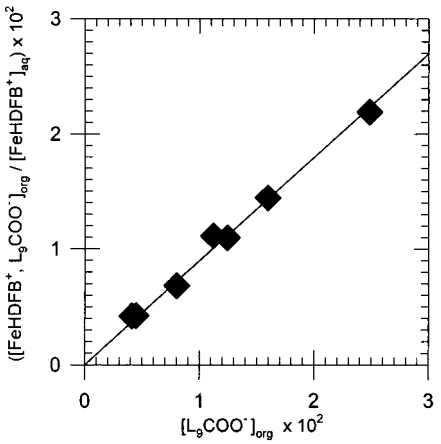


Figure 2. Plot of $[\{FeHDFB^+,L_9COO^-\}]_{org}/[FeHDFB^+]_{aq}$ as a function of $[L_9COO^-]_{org}$ according to eq 15 for the $FeHDFB^+$ extraction by L_9COO^- into chloroform at aqueous pH = 9.3, I = 0.1 ($FeHDFB^+$,Mg^{2+},ClO_4^-), $T = 25 \pm 0.1$ °C.

$COO^-\}]_{org}/[FeHDFB^+]_{aq}$ vs $[L_nCOO^-]_{org}$ at pH = 9.3 according to eq 15 yields a straight line for all three lariat ethers investigated. A representative plot for lariat ether **IIc** is shown in Figure 2. This establishes the validity of the 1:1 host−guest stoichiometry $\{FeHDFB^+,L_nCOO^-\}$ and confirms the anionic ionophore model, which assumes that the lariat ether carboxylic acid acts as both a host and counterion as shown in eq 14 and structure **III**. Equation 15 permits calculation of the extraction equilibrium constant (K_{ex}) at pH = 9.3 from the slope of the plot (Table 2).

(iv) Host−Guest Equilibria. The neutral ionophore model (eqs 1−3) may be used to represent the host−guest equilibrium at aqueous pH = 3.2 for supramolecular assembly formation involving ferrioxamine B and the undissociated lariat ether, as described by eqs 16 and 17. The equilibrium constant K,

$$\{FeHDFB^+,ClO_4^-\}_{org} + L_nCOOH_{org} \overset{K}{\rightleftharpoons} \{FeHDFB^+,L_nCOOH,ClO_4^-\}_{org} \quad (16)$$

$$K = [\{FeHDFB^+,L_nCOOH,ClO_4^-\}]_{org}/([\{FeHDFB^+,ClO_4^-\}]_{org}[L_nCOOH]_{org}) \quad (17)$$

defined in eqs 16 and 17, is the same as the K_a host−guest equilibrium constant defined for the crown ethers in our previous studies.[11−16] K is calculated according to eq 18 from extraction (K_{ex}, eq 13) and distribution (K_d, eq 7) data.

$$K = K_{ex}/K_d \quad (18)$$

The anionic ionophore model (eqs 4−6) may be used to represent the host−guest equilibrium at aqueous pH = 9.3, which leads to supramolecular assembly formation involving ferrioxamine B and the proton-dissociated lariat ether **III**, as represented by eqs 19 and 20. The host−guest association

$$FeHDFB^+_{org} + L_nCOO^-_{org} \overset{K_{app}}{\rightleftharpoons} \{FeHDFB^+,L_nCOO^-\}_{org} \quad (19)$$

$$K_{app} = [\{FeHDFB^+,L_nCOO^-\}]_{org}/([FeHDFB^+]_{org}[L_nCOO^-]_{org}) \quad (20)$$

Table 4. Host−Guest Association Equilibrium Constants for Ferrioxamine B ($FeHDFB^+$) with Crown Ether and Lariat Ether Carboxylic Acid Hosts in Chloroform at 25 ± 0.5 °C[a]

crown or lariat ether	log K,[b] pH = 3.2	log K_{app},[c] pH = 9.3
IIa, B18C6	3.21[d]	3.17[e]
IIb, L_6COOH	3.38	4.29
IIc, L_9COOH	3.18	4.55
IId, $L_{12}COOH$	3.26	4.85

[a] I = 0.1 ($FeHDFB^+$,Mg^{2+},ClO_4^-). [b] From eq 18. [c] From eq 21. [d] From Ref 13. [e] pH = 8.1.

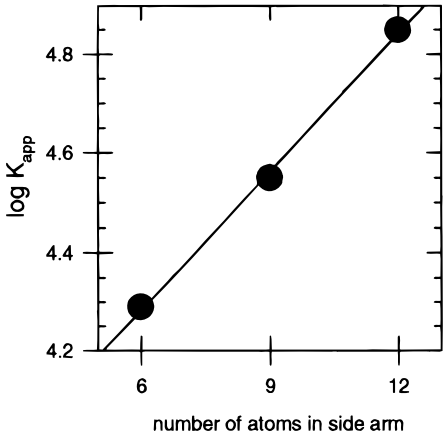


Figure 3. Plot of log K_{app} (eq 19) obtained at pH = 9.3 as a function of the lariat ether side arm length (number of atoms between the crown ether ring of the host and the carboxylic acid functional group).

constant K_{app} is calculated from eq 21, where D (eq 8) is the

$$K_{app} = K_{ex}/D \quad (21)$$

distribution of ferrioxamine B between the organic and aqueous phases at 0.08 M total perchlorate concentration and pH = 9.3. The extraction experiments at both pH = 3.2 and 9.3 are performed at the same total perchlorate concentration of 0.08 M. Therefore, the host−guest association constants K and K_{app} calculated by eqs 18 and 21, as given in Table 4, have the same units and can be readily compared.

Discussion

Comparison of the host−guest equilibrium constants for the lariat ethers at pH = 3.2 with those for the parent crown ether benzo-18-crown-6 (B18C6) allows us to determine the influence of the non-ionized side arm on the ability of the B18C6 cavity to act as a host for ferrioxamine B. The linearity of the plot shown in Figure 1 establishes that the lariat ethers form a three-component assembly $\{FeHDFB^+,L_nCOOH,ClO_4^-\}$ involving a counterion, as does the parent crown ether B18C6.[13] The data in Table 4 show that the non-ionized lariat ether side arm has no influence on assembly stability, as demonstrated by the similarity of the K values for the lariat ethers with that for B18C6.

At an elevated pH, the lariat ether serves as a host *and* a counteranion, to form an assembly $\{FeHDFB^+,L_nCOO^-\}$, as illustrated in **III**. This is demonstrated by the linearity of the plot in Figure 2. Comparison of K_{app} values for the lariat ethers with that for B18C6 at pH = 9.3 (Table 4) shows that the tethered, ionized carboxylate functional group significantly enhances host−guest complex stability. In addition, as shown in Figure 3, increasing the length of the side arm increases the stability of the assembly.

Steric factors are important in ferrioxamine B host−guest complex formation.[16] The stereochemical requirements of the H-bonding associated with host−guest complex formation, along with the bulkiness of ferrioxamine B, requires the N atom of the amine side chain to be well above the mean oxygen plane of the crown ether ring.[13,16] To assure charge neutrality, the carboxylate functionality is likely to be located close to the amine site through extensive bending of the side arm. The longer the arm, the better it is able to approach the distant amine site. It is highly improbable that ion pairing with the carboxylate functionality would overwhelm the electrostatic interactions of the amine site with the ether ring and thus pull the amine site of the metal complex away from the ether ring. Moreover, the complexation of ferrioxamine B in the crown ether cavity of the lariat ether is a combination of electrostatic and H-bonding interactions. If ferrioxamine B were in the $CHCl_3$ phase only because of ion pairing with the carboxylate functionality, then the K_{app} values should be the same for all of the lariat ethers, regardless of side arm length. This is clearly not the case and confirms that the lariat ethers are serving the dual function of host and counterion.

We have previously found that the variation in the stability of the {$FeHDFB^+$,DC18C6,X^-} assembly, where X^- is an external counterion, is related to the hydration enthalpy of both the cation and the anion.[12,15] The less negative the hydration enthalpy, the less the partial ion hydration in the organic phase and, therefore, the higher the stability of the assembly observed for the series of counterions X^- investigated. (Log K for {$FeHDFB^+$,DC18C6,X^-} increases with X^- in the sequence $Cl^- < NO_3^- <$ picrate $< ClO_4^-$.[15]) This raises the question as to whether the enhanced stability of the lariat ether relative to B18C6 at high pH is merely due to the presence of a carboxylate counteranion instead of ClO_4^-.

The carboxylate anion hydration enthalpy of -405 kJ mol^{-1} is much more negative than values reported for either picrate (-226 kJ mol^{-1}) or perchlorate (-209 kJ mol^{-1}).[44] This suggests, based on our previous studies,[12,15] that the host−guest association constant for the hypothetical ternary assembly {$FeHDFB^+$,B18C6,CH_3COO^-} (a good model for {$FeHDFB^+$, L_nCOO^-} in terms of hydration properties) should be much lower than that determined for {$FeHDFB^+$,B18C6,X^-} (X^- = pic^- or ClO_4^-).[13] However, the host−guest association constant for {$FeHDFB^+$,L_nCOO^-} obtained in this work is an order of magnitude higher than that for {$FeHDFB^+$,B18C6,ClO_4^-} (Table 4), which clearly shows that the unfavorable effect of carboxylate anion hydration is more than compensated for by an enhanced proximity effect by attachment to the crown ether. This is an effect which we may cautiously refer to as a "second-coordination-shell chelate effect".

Similarly, a change in anion hydration enthalpy is not responsible for the observed change in log K_{app} values with varying chain length lariat ethers (Figure 3). A simple calculation that includes additional $-CH_2-$ groups and their effect on total hydration enthalpy[45] leads to a possible change in log K_{app} of ca. 0.1 on going from L_6COO^- $L_{12}COO^-$. The observed change is much greater (ca. 0.6 log units; Table 4 and Figure 3), which suggests some additional effect (e.g., increased flexibility of the tether arm) that accounts for the differences in assembly stabilities with varying side arm length. The linearity of the plot in Figure 3 suggests that the host−guest complex stability may be enhanced even more by increasing the length of the carboxylate tether, presumably passing through a maximum at some optimal tether length.

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(44) Jazimirski, K. B. *Thermochemie von Komplexverbindungen;* Akademie-Verlag: Berlin, 1956.
(45) Vasil'ev, V. P.; Zolotarev, E. K.; Kaustinskii, A. F.; Mishchenko, K. P.; Podgornaya, E. A.; Jazimirski, K. B. *Zh. Fiz. Khim.* **196**0, *34*, 1763; *Russ. J. Phys. Chem.* (Engl. transl.) **196**0, *34*, 840.