Journal of Alloys and Compounds 303–304 (2000) 112–115
www.elsevier.com/locate/jallcom

Eu(III) complexation constants with glutamine and serine in aqueous methanol

Herbert B. Silber*, Nastaran Ghajari, Victor Maraschin

Department of Chemistry and Nuclear Science Facility, San Jose State University (SJSU), San Jose, CA 95192-0101, USA

Abstract

A problem of long-term interest in lanthanide chemistry is whether a ligand resides in the cation inner or outer solvation shell. We have been using deviations from Beer's Law on hypersensitive peaks to determine the complexation constants as a function of temperature in water and mixed solvents via the Benesi–Hildebrand method. In our previous studies using alanine and glycine, as well as in this investigation, only a single complex forms. Enthalpy and entropy data are used to predict if a complex is outer or inner sphere in water. These results will be compared to other lanthanide complexation systems.

Keywords: Europium complexes; Serine; Glutamine; Inner sphere complexes; Outer sphere complexes

1. Introduction

Investigations of the complexation between lanthanide ions and ligands in aqueous organic solvents have provided information about lanthanide coordination number changes and the nature of the species as inner or outer sphere complexes [1,2]. In recent years, the substitution of Eu(III) for calcium and magnesium has resulted in lanthanide probes in biological systems [3]. Information about how many water molecules remain in the inner coordination shell of Eu(III) complexes can be used to examine metal ion binding to biological molecules by the luminescence lifetimes method using the Horrocks equation [4]. We have been investigating the complexation between Eu(III) and amino acids in aqueous methanol in an attempt the understand the binding of Eu(III) to these molecules of biochemical interest [5,6]. Based upon Choppin's thermodynamic model, which predicts that inner sphere complexes have positive enthalpies and entropies and that outer sphere complexes have negative enthalpies and entropies [7], Eu(III) with glycine was predicted to be inner sphere [5]. This result was confirmed by the use of ultrasonic relaxation methods [5]. However, alanine [8] had negative enthalpy and entropy of complexation in water and appeared to be outer sphere. Glutamine and serine complexes with Eu(III) in water were similar to the glycine system and were predicted to be inner sphere [9]. Other differences were observed between alanine and glycine complexes as methanol was added to the solvent system [6] and this investigation of the solvent dependence of the complexation between Eu(III) and glutamine and serine was initiated.

2. Experimental

After dissolving dried Eu_2O_3 (Molycorp) in a slight excess of $HClO_4$, the stock solutions were standardized by EDTA titrations [5]. L-Serine (99%, Sigma) and L-glutamine (99%, Sigma) were used without further purification. Our previous studies with alanine and glycine had demonstrated that no proton is released upon complexation with the zwitterion in our pH range, and thus complexation does not change the ionic strength (1.0 M in perchlorate media) in 0.167 M $Eu(ClO_4)_3$ with small quantities of $HClO_4$ or NaOH to adjust the pH. Thermostated Cary 3 spectrometers were used to record the absorbance for each solution at 15, 20, 25 and 37°C. At constant europium concentration the addition of either amino acid to the solution increases the absorbance, a feature absent when excess non-coordinating $NaClO_4$ was added. The absorption increase is attributed to complexation between Eu(III) and the amino acids and the small increase in absorption is attributed to a higher extinction coefficient for the complex than for the free Eu(III) ion. Measurements were carried out between 250 and 500 nm, with absorption at 464.8 nm used to determine the complexation constants. In each solvent mixture, the absorbance was investigated at con-

---

*Corresponding author.

0925-8388/00/$ – see front matter © 2000 Elsevier Science S.A. All rights reserved.
PII: S0925-8388(00)00614-9

stant metal ion and amino acid concentration as a function of operational pH reading to determine the experimental region where absorption is independent of concentration. No attempt was made to correlate the pH meter reading with real solution acidities in aqueous methanol since we work in a region where the complexation is pH independent. For one-to-one complexes, the Benesi–Hildebrand method has been useful in determining the complexation constants [10]. Alternate forms of this expression can be derived for the highest complex in a series [11]. Difference spectra between the test solution containing the Eu(III), the amino acid and the solvent versus the reference solution containing the metal ion, ionic media, and the same solvent as the test solution are obtained using matched 1.00-cm cells (Helma). The Benesi–Hildebrand equation is:

$$1/\Delta A = (1/M_o K\Delta\varepsilon L_o) + (1/M_o\Delta\varepsilon) \tag{1}$$

with ΔA as the absorbance difference between the two Eu(III) solutions; $\Delta\varepsilon$ is the extinction coefficient difference between them; M_o is the total Eu(III) concentration which is constant in these experiments; and L_o is the total ligand concentration. Thus, a plot of $1/\Delta A$ as a function of $1/L_o$ gives a straight line with an intercept equal to $(1/M_o\Delta\varepsilon)$ and a slope equal to $(1/M_o K\Delta\varepsilon)$, thereby allowing the calculation of the complexation constant by dividing the intercept by the slope. We attempted to refine the calculation by using our calculated equilibrium constant to re-determine L_o, and then re-plot the data according to Eq. (1). This attempt failed in that a negative intercept was found and the only parameter in Eq. (1) which could be negative is $\Delta\varepsilon$ which would require that the extinction coefficient for the complex is lower than that of free metal, and this fails for two reasons: (1) this result is contrary to experiment where the absorbance increases as complex forms; and (2) the slope would also have to be negative, which was also not observed.

3. Results and discussion

The first experiments done at each solvent composition were to determine Beer's Law for $Eu(ClO_4)_3$. At each solvent composition and temperature, Beer's Law was observed for the $Eu(ClO_4)_3$ even at absorbancies greater than one. Second, for each solvent composition it is necessary to determine a range where the absorbance is independent of the pH and in water this is between pH 2 and about 5 and is an actual pH. However, in aqueous methanol the pH meter reading loses its significance other than as an operational parameter to indicate where the absorbance measurements are reproducible.

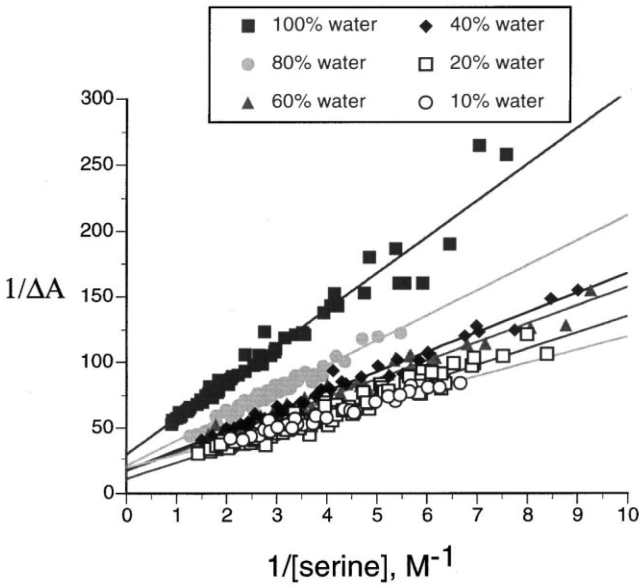

Fig. 1. Benesi–Hildebrand data for 0.167 M Eu(III) with serine at 25°C at 464.8 nm and an ionic strength of 1.0 ($NaClO_4$).

The Benesi–Hildebrand data are linear at each solvent composition and temperature. Fig. 1 is a sample plot of the data for Eu(III) with serine at 25°C. The stability constants were obtained for both amino acid systems and are summarized in Table 1. The variation of equilibrium constants with temperature, called the van't Hoff equation, can be used to calculate the entropy and enthalpy of complexation by the following equations:

$$\Delta G^o = -RT \ln K = \Delta H^o - T\Delta S^o \tag{2}$$

which can be rearranged to give:

$$\ln K = -(\Delta H^o/RT) + \Delta S^o/R \tag{3}$$

and a graph of $\ln K$ as a function of the inverse absolute temperature gives a straight line of slope $-\Delta H^o/R$ and an intercept of $\Delta S^o/R$. For each system, the van't Hoff plots were used to determine the complexation enthalpy and entropy, and a typical result is shown in Fig. 2. These results are summarized in Table 2 and Figs. 3 and 4.

Table 1
Equilibrium constants for the formation of the Eu(III)–amino acid complexes as a function of solvent and temperature

Percent water	15°C	20°C	25°C	37°C
Europium(III)–serine				
100	0.87±0.12	0.91±0.10	1.02±0.12	1.07±.010
80	1.08±0.15	1.01±0.14	1.03±0.12	0.97±0.12
60	0.92±0.12	1.04±0.10	1.08±0.12	1.10±.010
40	0.97±0.22	1.09±0.21	1.10±0.18	1.16±0.21
20	0.72±0.11	0.77±0.11	0.76±0.11	0.86±0.12
10	1.83±0.21	1.77±0.23	1.80±0.23	3.00±.48
Europium(III)–glutamine				
100	1.11±0.24	1.34±0.23	1.51±0.17	1.67±0.23
80	1.00±0.08	1.14±0.11	0.98±0.12	1.09±0.13
60	0.58±0.10	0.54±0.11	0.60±0.11	0.59±0.12
40	0.83±0.11	0.71±0.11	0.73±0.17	0.50±0.10
20	0.53±0.08	0.60±0.10	0.60±0.10	0.60±0.12
10	0.54±0.12	0.64±0.12	0.85±0.11	0.91±0.15

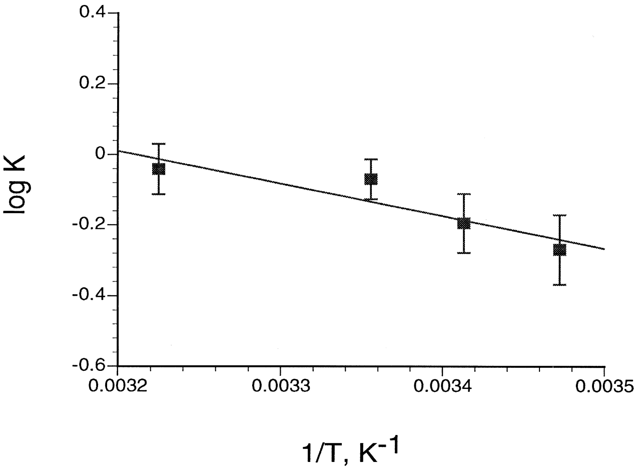

Fig. 2. van't Hoff plot for Eu(III) with glutamine in 10% water/90% methanol.

Table 2
Complexation enthalpy and entropy for the Eu(III)–amino acid complexes as a function of percent water

Percent water	Serine enthalpy (kJ/mol)	Serine entropy (J/mol-K)	Glutamine enthalpy (kJ/mol)	Glutamine entropy (J/mol-K)
100	6.8±1.7	23±6	13±4	47±12
80	−3.1±1.1	−10±4	1.4±16	5±5
60	5.6±2.4	−19±8	2.0±2.2	2±7
40	5.2±2.1	18±7	−7.1±3	−25±10
20	6.8±1.5	21±5	2.4±2.7	5±9
10	18±7	66±23	18±6	57±19

Because the absorbance errors are relatively large and the complexation equilibrium constants are relatively small, the results have some scatter. The reported errors are those directly obtained by the least-squares analysis. Although the absolute errors reported look low, the percent error in the enthalpy and entropy values range from 22 to over 1000%. In lanthanide systems, it is not uncommon for the enthalpy and entropy terms to follow the same general relationship. In our system where the equilibrium constants are near unity this is even more evident since mathematically ΔH° and $T\Delta S^\circ$ must be almost equivalent when ΔG° is near zero. Even though it has been hypothesized that the entropy and enthalpy nearly compensate for each other in many lanthanide equilibrium systems, the entropies were calculated from the intercept. The glutamine result in 40% water originally was calculated to have both a more negative slope and intercept. Thus, the addition of 16 new metal/ligand compositions were studied and made an average difference in the equilibrium constants of ±0.03, a result which was well within the calculated error limits. These revised values are included in the tables and figures. The presence of both a negative enthalpy and entropy for the 40% water system is difficult to explain in terms of inner versus outer sphere complexes. In the Eu(III) alanine system, a small maximum in enthalpy was observed at 80% water, and then the enthalpy decreased as more methanol was present in the solvent [6], and this maximum is absent for both amino acids in this study. In aqueous methanol within the solvent range studied, the preference of europium for water over methanol in the inner solvation shell is well known, and should not be different with a difference in these amino acids. As methanol is added, it is expected that the relative concentrations of inner sphere complex will increase, but also outer sphere complexes may also form. Thus, it may well be that the thermodynamic differences observed might reflect varying ratios of outer sphere to inner sphere complex in these solvated Eu(III) ions and this ratio is a function of the ligand.

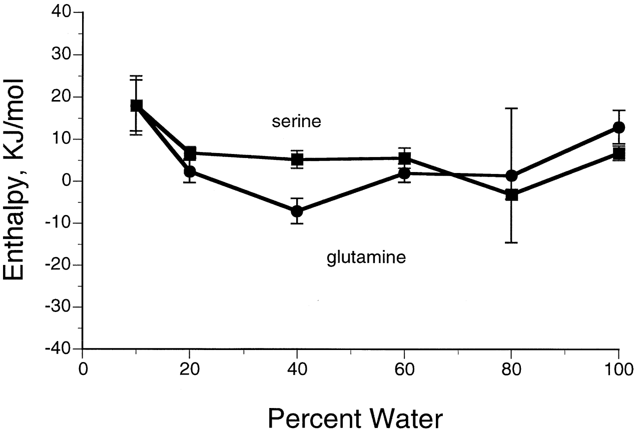

Fig. 3. Complexation enthalpy for Eu(III) with serine and glutamine.

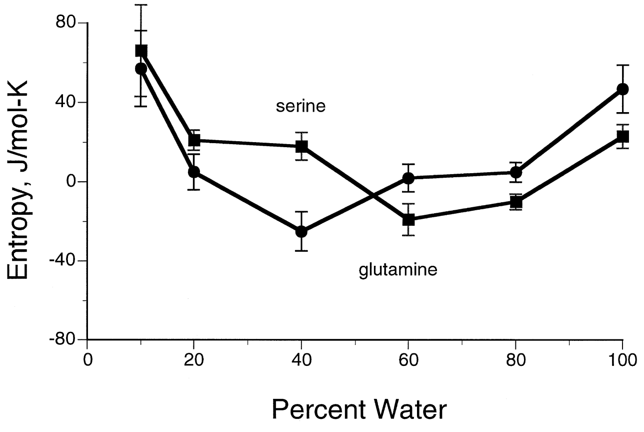

Fig. 4. Complexation entropy for Eu(III) with serine and glutamine.

Fig. 5 compares the results of this study with previous systems. The following conclusions can be drawn: the complexation enthalpy and entropy in water are consistent with predominantly inner sphere complexes between Eu(III) and simple amino acids. It is interesting that all four amino acid systems have similar complexation constants in water. Except for the glycine system, the addition of methanol, a solvent with a lower dielectric constant than water has relatively little effect upon the complexation. This is consistent with our hypothesis that complexation of the Eu(III) is with the neutral amino acid, since the overall

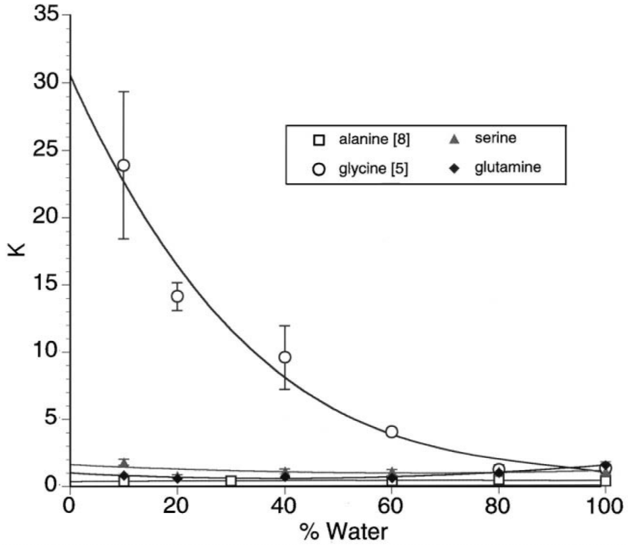

Fig. 5. Complexation constants for Eu(III) with amino acids at 25°C and 1.0 M ionic strength ($NaClO_4$).

charge on the Eu(III) complex remains the same as free solvated Eu(III), the solvent dielectric constant change should not be as important as in systems where complexation reduces the charge of the reacting species. Because the amino acids are zwitterions with a positive charge on the amino groups, it is not surprising that the magnitude of the complexation constants are significantly lower than those observed when the ligands have overall negative charges.

Acknowledgements

We would like to acknowledge the financial support of the Petroleum Research Fund administered through the American Chemical Society Grant #32100-B3 and the National Institutes of Health Minority Biomedical Research Support (MBRS) Program Grant 2 S06 GM08192 for the support for this research.

References

[1] H.B. Silber, D. Bouler, T. White, J. Phys. Chem. 82 (1978) 775.
[2] H.B. Silber, M. Strozier, Inorg. Chim. Acta 128 (1987) 267.
[3] J.-C.G. Bunzli, G.R. Choppin, in: Lanthanide Probes in Life, Chemical and Earth Sciences, Elsevier, Amsterdam, 1989.
[4] W.D. Horrocks Jr., A. Albin, Prog. Inorg. Chem. 31 (1984) 1.
[5] H.B. Silber, R.L. Campbell, N. Nguyen, T. Parker, S.P. Sibley, J. Alloys Comp. 225 (1995) 291.
[6] H.B. Silber, Y. Nguyen, J. Alloys Comp. 275–277 (1998) 811.
[7] G.R. Choppin, Coord. Chem. Rev. 18 (1976) 199.
[8] H.B. Silber, Y. Nguyen, R.L. Campbell, J. Alloys Comp. 249 (1997) 99.
[9] H.B. Silber, N. Ghajari, V. Maraschin, Mater. Sci. Forum 315–317 (1999) 490.
[10] H.A. Benesi, J.H. Hildebrand, J. Am. Chem. Soc. 71 (1949) 2705.
[11] H.B. Silber, M.A. Murguia, Inorg. Chem. 24 (1985) 3794.