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Research Article

Stability constant of oxovanadium(IV)-levofloxacin complex in pure and binary mixed solvents

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ABSTRACT

The complexation between the oxovanadium(IV) ion and the levofloxacin (levH) was examined in pure solvents (water; H_2O, dimethylformamide; DMF, and methanol; MeOH) and mixed solvents (dimethylformamide-water; DMF-H_2O and methanol-water; MeOH-H_2O) using the spectrophotometric method. The complexation reaction was also observed in aqueous solution (pH 2) at various ionic strengths and temperatures. The results indicate that the complex has a 1:2 stoichiometry in all pure and binary mixed solvents. The Harvey and Manning equation was used to calculate the stability constant (logβ) of the oxovanadium(IV)-levofloxacin [$V^{IV}O(lev)_2$] complex. In pure solvents, the logβ values of the [$V^{IV}O(lev)_2$] complex follow the order MeOH > H_2O > DMF. The MeOH-H_2O system has substantially larger logβ values than the DMF-H_2O system. Changes in logβ versus the dielectric constant (1/D) of binary mixed solvents show a linear relationship. As ionic strength and temperature increase in aqueous solution, the stability constant of the [$V^{IV}O(lev)_2$] complex decreases. Based on the thermodynamic parameters (ΔG, ΔH, and ΔS), the complexation reaction between the oxovanadium(IV) cation and levofloxacin is spontaneous and exothermic.

Introduction

Levofloxacin (levH; Fig. 1) is a third-generation fluoroquinolone antibiotic that inhibits bacterial multiplication by preventing the replication and repair of bacterial DNA (Das et al., 2014). The carbonyl and carboxylic acid functionalities of quinolones provide effective sites for chelation with divalent or trivalent metal ions (Sultana et al., 2013). Quinolones can form 1:1, 2:1, and 3:1 chelates, depending on the specific metal ion, the relative quinolone concentration, and the pH of the reaction medium (Sultana et al., 2013). According to reports, neutral quinolones in the zwitterionic state can form simple bidentate complexes (Sultana et al., 2013).

According to the literature, metal complexes of quinolone molecules play an important role in their biological activities (Sultana et al., 2013; Drevensek et al., 2006; Li et al., 2008; Vieira et al., 2009; Gouvea et al., 2012; Patel et al., 2012; Tarushi et al., 2011; Valentina, 2013; Bhardwaj and Singh, 2014; Sabale et al., 2012; Urbaniak and Kokot, 2013; Mehta et al., 2014; Sultana et al., 2014; Sabale et al., 2012). In 2013, Sultana et al. synthesized levofloxacin-metal (Mn^{II}, Co^{II}, Ni^{II}, Cu^{II}, and Zn^{II}) complexes and evaluated their antimicrobial activity against eleven microorganisms. The immune response to these complexes has also been evaluated. Drevensek et al. (2006) examined the complexation of magnesium

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with levofloxacin and ofloxacin, whereas Li et al. (2008) investigated the complexes of levofloxacin and ofloxacin with copper in aqueous solution. Palladium and platinum complexes of levofloxacin exhibited antitubercular activity (Vieira et al., 2009), whereas gold(III)-levofloxacin exhibited cytotoxic activity against A20 (murine lymphoma), B16-F10 (murine melanoma), and K562 (human myeloid leukemia) tumor cell lines (Gouvea et al., 2012).

However, the stoichiometry and binding constant of the oxovanadium(IV)-levofloxacin complex in various solvents remain uncharacterized. Recently, we found that the oxovanadium(IV) ion coordinates with ciprofloxacin (cfH) to form the $[V^{IV}O(cf)_2]$ complex in aqueous, organic, and aqueous-organic mixed solvents (Shaon et al., 2022). A literature survey indicates that the $[V^{IV}O(cf)_2]$ complex exhibits potent insulin-mimetic activity (Rehder et al., 2002). These findings motivated us to investigate the stoichiometry and binding constant of the oxovanadium(IV)–levofloxacin complex in aqueous, organic, and aqueous–organic mixed solvents to better understand its complexation behavior in solution.

The complexation of levofloxacin with the oxovanadium(IV) ion was studied spectroscopically in pure (deionized water, methanol, and dimethylformamide) and mixed (methanol-water and dimethylformamide-water) solvents. In each solvent, the stoichiometry and stability constant of the oxovanadium(IV)-levofloxacin complex were determined. In aqueous solution, the stability constant of the oxovanadium(IV)-levofloxacin complex was measured at various ionic strengths and temperatures.

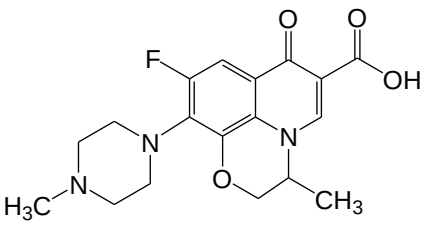

Fig. 1. The structure of levofloxacin

Materials and Methods

The levofloxacin (levH) ligand was obtained from Suprime Pharmaceutical Ltd., Dhaka, Bangladesh. The oxovanadium salt ($VOSO_4$) was obtained from Wako Pure Chemical Industries, Ltd., Japan. The sodium salt (Na_2SO_4) was purchased from Merck (India) Limited, Mumbai-400018, India, and used without further purification. This investigation utilized pure solvents, such as deionized water (H_2O), methanol (MeOH), and dimethylformamide (DMF), as well as mixed solvents, such as MeOH-H_2O and DMF-H_2O (w/w). Reagent-grade MeOH and DMF were purified by standard distillation. Deionized water was produced by passing distilled water through a deionization column (Barnstead, Sybron Corporation, USA).

To determine the stoichiometry of the oxovanadium(IV)-levofloxacin complex in solution, the molar-ratio method (Yoe and Jones, 1944) was used. In this method, the metal-ion concentration (5×10^{-6} M) was held constant while the ligand concentration ($2.5–30\times10^{-6}$ M) was varied. The pH of the samples was adjusted by adding microliter quantities of 2 M H_2SO_4. The solution pH was measured with a pH meter (Adwa AD 8000). When the absorbance of the oxovanadium(IV) complex was plotted against the molar ratio of ligand to metal (L:M), two straight lines were obtained, and their intersection revealed the complex's stoichiometry. Using the following Equation (1) proposed by Harvey and Manning (1950), the apparent global stability constant (β) of the oxovanadium(IV)-levofloxacin complex in solution was calculated:

$$\beta = \frac{A/A_m}{\left(1 - A/A_m\right)^{n+1} C_L{}^n n^2} \qquad (1)$$

where A is the absorbance at the ligand concentration C_L, A_m is the absorbance after complete reaction, when all metal ions have reacted with excess ligand, and n is the complex's stoichiometry. The absorbance (A_m)

for complete complexation was determined by measuring the absorbance of a solution containing a 20-fold excess of the ligand, assuming that all metal ions were present as metal-ligand complexes.

The UV-visible absorption spectra were recorded with a Shimadzu UV-1800 spectrophotometer equipped with an electronically thermostated cell holder (Shimadzu) in a quartz cell with a 1 cm path length. The temperature was monitored with a copper-constantan thermocouple attached to the quartz cell. Before each measurement, the spectrophotometer's reference line was calibrated against the solvent used for the respective solution.

To examine the effects of ionic strength and temperature on oxovanadium(IV)-levofloxacin complexation, the stability constant of the complex was measured over a range of ionic strengths (4.5-6.0)×10^{-3} M and temperatures 30-45°C (±0.2 °C) in aqueous solution. The ionic strength of the reaction medium was adjusted by adding solid Na_2SO_4 to the metal-ligand solution in the required amounts. Using the law of addition, the dielectric constant (*D*) values of binary mixtures were calculated (Shaon et al., 2022).

Results and Discussion

Fig. 2 shows the UV absorption spectra of oxovanadium(IV) sulfate ($VOSO_4$), levofloxacin (levH), and the oxovanadium(IV)-levofloxacin complex in aqueous solution (pH 2). The complexation between $VOSO_4$ and levH occurred in aqueous solution at pH 2 to prevent the oxidation of oxovanadium(IV) ion at higher pH (Karmaker et al., 2006). Fig. 2a shows that free $VOSO_4$ exhibits no UV absorption maximum. UV absorption spectra (Fig. 2b) reveal distinct absorption maxima (λ_{max}) at 289 and 330 nm for free levH and at 296 and 324 nm for the oxovanadium(IV)-levofloxacin complex. Therefore, the 7-nm red shift of the absorption band near 296 nm and the 6-nm blue shift of the absorption band at 324 nm in the UV region must be attributed to the electronic transition associated with the formation of the complex (Shaon et al., 2022).

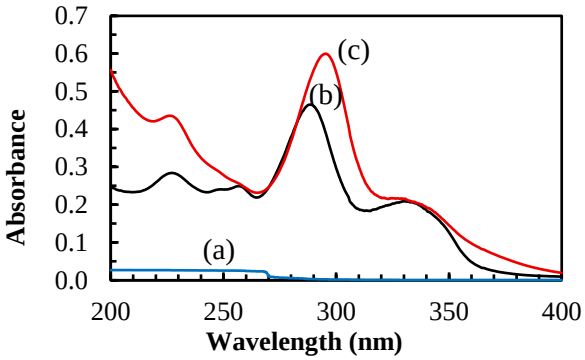

Fig. 2. Typical UV absorption spectra of free metal ion (a: $VOSO_4$), free ligand (b: levH), and complex (c: $[V^{IV}O(lev)_2]$) in aqueous solution at pH 2.

Fig. 3a shows the absorption maximum (λ_{max}) at 762 nm and a shoulder at 623 nm in the visible spectrum of $VOSO_4$ in an aqueous solution (pH 2). Free levH exhibits no absorption maximum in the visible spectrum (Fig. 3b). Fig. 3c depicts the absorption maximum (λ_{max}) of the oxovanadium(IV)-levofloxacin complex in an aqueous solution (pH 2) at approximately 814 nm, with a shoulder at 606 nm. Thus, the red (52 nm) and blue (17 nm) shifts of two typical bands at 814 and 606 nm, respectively, indicate that the oxovanadium(IV) metal ion is coordinated by levH. Similar outcomes were observed in the aqueous absorption spectra of oxovanadium(IV)-ciprofloxacin (Shaon et al., 2022), oxovanadium(IV)-poly-γ-glutamic acid (Karmaker et al., 2006), and oxovanadium(IV)-suprofen complexes (Williams et al., 1998).

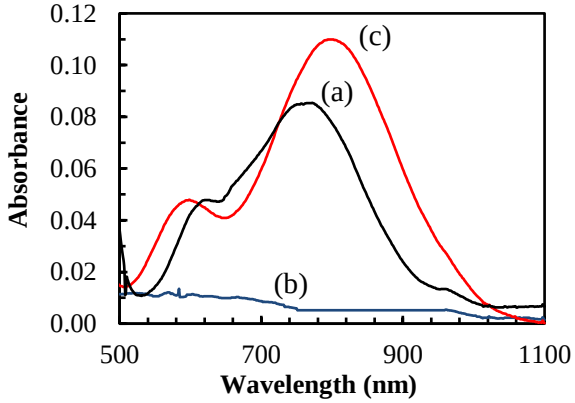

Fig. 3. Typical visible absorption spectra of free metal ion (a: $VOSO_4$), free ligand (b: cfH), and complex (c: $[VO(IV)(cf)_2]$) in aqueous solution at pH 2.

Due to the limited solubility of $VOSO_4$ and the oxovanadium(IV)-levofloxacin complex in MeOH and DMF, only UV absorption spectra were measured for free $VOSO_4$, free levH, and the oxovanadium(IV)-levofloxacin complex. Figs. 4a and 5 show no absorption maxima for $VOSO_4$ in MeOH and DMF, respectively. The UV absorption spectra of free levH revealed distinct absorption maxima (λ_{max}) at 292 and 330 nm in pure MeOH (Fig. 4b) and at 302 and 326 nm in pure DMF (Fig. 5b). The absorption maxima (λ_{max}) of the oxovanadium(IV)-levofloxacin complex were measured at 297 and 326 nm in MeOH (Fig. 4c) and at 297 and 329 nm in DMF (Fig. 5c). Consequently, the blue shift at shorter wavelengths (326 nm in MeOH and 297 nm in DMF) and the red shift at longer wavelengths (297 nm in MeOH and 329 nm in DMF) must be attributed to electronic transitions associated with the formation of the complex.

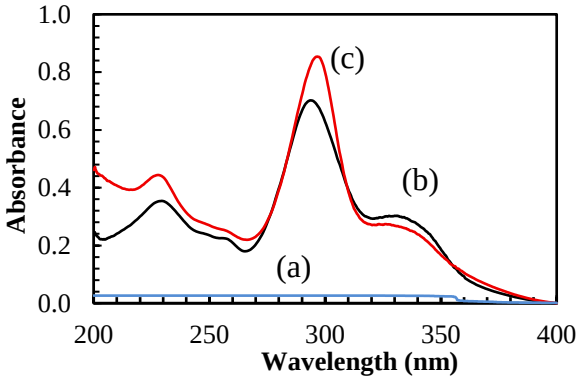

Fig. 4. Typical UV absorption spectra of free metal ion (a: $VOSO_4$), free ligand (b: levH), and complex (c: $[V^{IV}O(lev)_2]$) in MeOH.

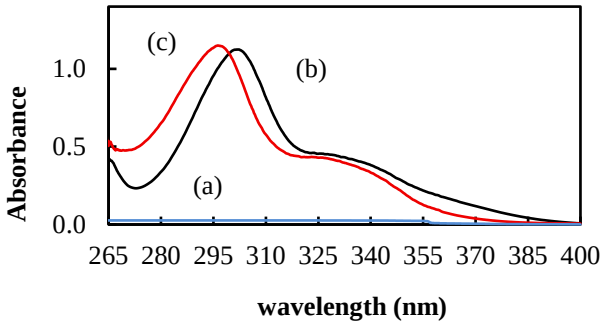

Fig. 5. Typical UV absorption spectra of free metal ion (a: $VOSO_4$), free ligand (b: levH), and complex (c: $[V^{IV}O(lev)_2]$) in DMF.

The absorption maxima of the oxovanadium(IV)-levofloxacin complex shifted from 297 and 329 nm in DMF to 296 and 324 nm in aqueous solution. The complex's peak values followed the order: DMF > MeOH > H_2O. Except for DMF, this sequence aligns with the typical pattern of spectral band blue shifts as solvent polarity increases. This is due to DMF's exceptionally strong electron-donating properties (Gutmann, 1978). The donor numbers of the pure solvents are: DMF > MeOH > H_2O. Their dielectric constants follow the order: MeOH < DMF < H_2O. Generally, a polar solvent stabilizes the π, π*, and n orbitals through solvation, with the π* orbital being more stabilized than the π orbital. Likely because the π* orbital is more polar, the π → π* transition diminishes with solvation, causing a red shift, while the n → π* transition increases, resulting in a blue shift. As solvent polarity increases from MeOH to DMF to H_2O and the alkyl chain length decreases, the maximum absorption wavelength shifts to lower values.

Fig. 6 shows the formation curve of the oxovanadium(IV)-levofloxacin complex in aqueous solution using the molar ratio method at 324 nm. The stoichiometry of the oxovanadium(IV)-levofloxacin complex was determined to be 1:2 (Fig. 6). The constancy of the absorbance reading indicates that only one complex was formed under experimental conditions. Analogous results were observed in oxovanadium(IV)-ciprofloxacin (Shaon et al. 2022), iron(III)-sulfosalicylate (Karmaker et al., 2003), and iron(III)-salicylate (Karmaker et al. 2004) complexes in aqueous solutions. Similar formation trajectories (figure not shown) were observed in MeOH and DMF. According to these results, only the $[V^{IV}O(lev)_2]$ complex was formed in H_2O, MeOH, and DMF. Using the formula described in the experimental section, the apparent global stability constant (β) of the $[V^{IV}O(lev)_2]$ complex was determined twice in solution at the respective absorption maxima (λ_{max}). Table 1 displays the average logβ value for $[V^{IV}O(lev)_2]$ in solution.

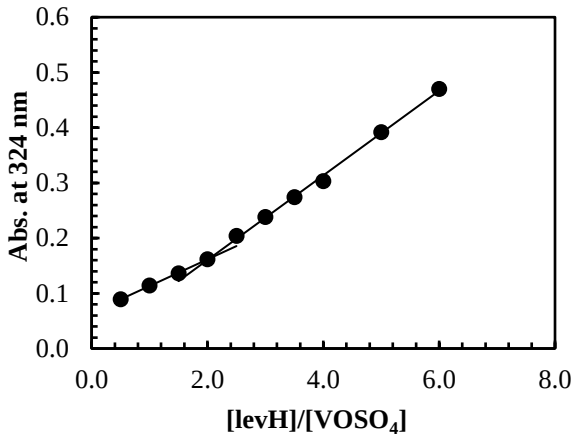

Fig. 6. Typical formation curves of oxovanadium(IV)-levofloxacin in aqueous solution (pH 2).

The concentration of metal ion (5×10^{-6} M) was kept constant while that of the ligand ($2.5–30\times10^{-6}$ M) was regularly varied in the molar ratio method.

The order of stability of the $[V^{IV}O(lev)_2]$ complex in pure solvents is MeOH>H_2O>DMF. This sequence is consistent with the natural order of stability of complexes and with the polarity and dielectric constant of solvents, except for DMF. The formation of complexes involves replacing metal-solvent bonds with metal-ligand bonds; however, one of DMF's resonance structures hinders the formation of metal-ligand bonds, thereby decreasing the stability constant (Karmaker et al., 2004). In addition, the observed outcomes can be explained in terms of the donor-acceptor properties of solvents. In general, a strong donor solvent (DN_{DMF} = 26.6 > DN_{MeOH} = 19.0 > DN_{water} = 18.0) promotes dissociation of the complex via coordination to the metal ions, thereby decreasing the complex's stability constant (Gutmann, 1978). Similar results were observed for the oxovanadium(IV)-ciprofloxacin complex in solutions (Shaon et al. 2022).

The values of logβ (Table 1) for the $[V^{IV}O(lev)_2]$ complex increased with increasing DMF and MeOH concentrations in DMF-H_2O and MeOH-H_2O binary mixtures, respectively. For a given binary mixture composition, the value of logβ for the $[V^{IV}O(lev)_2]$ complex is greater in the MeOH-H_2O system than in the DMF-H_2O system. This may be due to the decrease in the dielectric constant and donor number of the MeOH-H_2O binary mixture (Gutmann 1978).

Table 1. Stability constant of the $[V^{IV}O(lev)_2]$ complex in different pure and binary mixed solvents at 30°C.

Solvent	Dielectric constant	Stability constant (logβ)
H_2O	78.4	8.66
DMF	36.7	8.63
MeOH	32.6	8.72
DMF–H_2O binary mixed solvent (% w/w)		
10:90	74.23	8.35
20:80	70.06	8.38
30:70	65.89	8.41
40:60	61.72	8.44
50:50	57.55	8.46
60:40	53.38	8.48
70:30	49.21	8.52
80:20	45.04	8.56
90:10	40.87	8.60
MeOH–H_2O binary mixed solvent (% w/w)		
10:90	73.89	8.45
20:80	69.28	8.48
30:70	64.69	8.50
40:60	60.12	8.52
50:50	55.55	8.54
60:40	50.98	8.58
70:30	46.34	8.60
80:20	41.76	8.63
90:10	37.18	8.68

This tendency was also observed in complexes containing oxovanadium(IV)-ciprofloxacin (Shaon et al., 2022), iron(III)-sulfosalicylate (Karmaker et al.,

2003), and iron(III)-salicylate (Karmaker et al., 2004).

When logβ was plotted against 1/*D* (Fig. 7), straight-line relationships were observed, with distinct slopes and intercepts for the DMF-H_2O and MeO-H_2O binary systems. Two linear equations were derived using least squares analysis:

DMF-H_2O :

$\log\beta = 22.027(1/D) + 8.0744$, $n = 10$, $R^2=0.9837$

MeOH-H_2O :

$\log\beta = 15.489(1/D) + 8.2589$, $n = 10$, $R^2=0.9868$

These results suggest that the acid-base properties of the solvents and ion-solvent interactions play a significant role in determining the stability of the $[V^{IV}O(lev)_2]$ complex in solution (Karmaker et al., 2004).

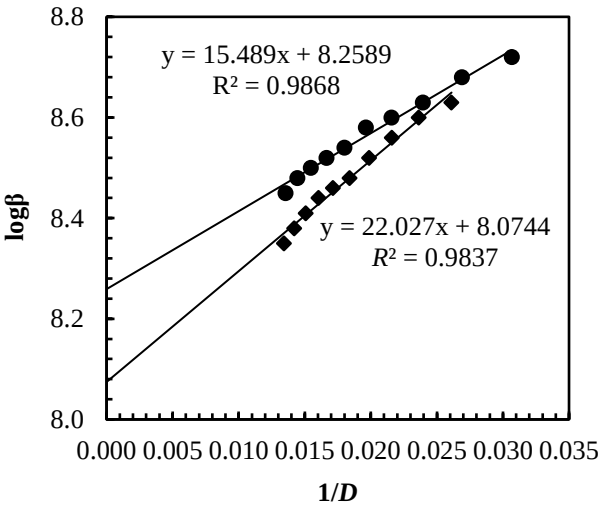

Fig. 7. Plot of log*β* vs. 1/*D* for $[V^{IV}O(lev)_2]$ complex in DMF-H_2O (♦) and in MeOH-H_2O (●) binary mixed solvents.

In aqueous solution, the log*β* values for the $[V^{IV}O(lev)_2]$ complexes were estimated at various ionic strengths and temperatures (Table 2). Table 2 also presents the thermodynamic parameters for the metal-ligand complexation reaction in aqueous solution.

Table 2. Stability constants of $[V^{IV}O(lev)_2]$ complex at different ionic strengths and temperatures in aqueous solution (pH 2) and thermodynamic parameters of the complexation reaction.

Parameter					
Ionic strength (*I*) M	0.0000	0.0045	0.0050	0.0055	0.0060
Stability Constant (log*β*)	9.82	8.64	8.59	8.52	8.47
Temperature (*T*) °C		30	35	40	45
Stability Constant (log*β*)		8.66	8.60	8.56	8.52
Thermodynamic parameters					
Δ*G* (kJ/mol)	−56.97	−50.24	−50.72	−51.30	−51.88
Δ*H* (kJ/mol)			−16.66		
Δ*S* (J/K/mol)			110.76		

The effect of ionic strength on the $[V^{IV}O(lev)_2]$ complex was studied only in aqueous solution at constant temperature, using various concentrations of salt (Na_2SO_4). The ionic strength of the reaction medium had a significant effect on the stability constant of the $[V^{IV}O(lev)_2]$ complex. At constant temperature, the values of log*β* (Table 2) decreased progressively as the ionic strength of the aqueous solution increased. These results suggest that the $[V^{IV}O(lev)_2]$ complex forms through the reaction of positively charged metal ions with a negatively charged ligand (Shaon et al., 2022; Karmaker et al., 2004). The thermodynamic metal-ligand stability constant, $\log\beta_{I=0}$, was derived from the plot of logβ versus $\sqrt{I}$ by extrapolating to zero ionic strength (Figure not given; R^2=0.996). I=0 denotes that no ionic salt was applied at that time.

As the solution temperature increased, the metal-ligand stability constant (logβ; Table 2) of the $[V^{IV}O(lev)_2]$ complex in aqueous solution decreased. These results indicate that the $[V^{IV}O(lev)_2]$ complexation is exothermic. Using conventional equations, the thermodynamic parameters for the change in free energy (ΔG), enthalpy (ΔH), and entropy (ΔS) were determined as follows:

$$\Delta G = -2.303RT \log\beta$$

$$\log\beta = -\Delta H/2.303RT + \Delta S/2.303R$$

where logβ represents the stability constant of the $[V^{IV}O(lev)_2]$ complex, T presents the solution temperature (K), and R represents the gas constant (8.314 J/mol.K). The values of ΔH and ΔS were derived from the slope and intercept of the Van't Hoff plot of logβ versus 1/T (Fig. 8).

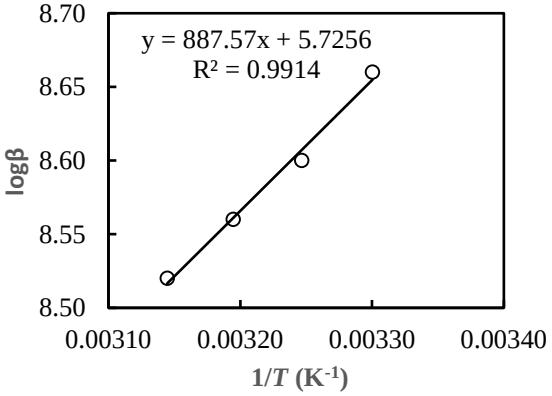

Fig. 8. Plot of logβ vs. 1/T for $[V^{IV}O(lev)_2]$ complex in aqueous solution (pH 2).

The results are shown in Table 2. The negative free energy change indicates that the entire system is spontaneous from a thermodynamic standpoint. The negative value of ΔH confirms that the complexation reaction is exothermic. The positive value of ΔS indicates that the complex formation reaction is entropically favorable. The larger ΔS value indicates that the complex has a 1:2 stoichiometry.

Conclusion

The absorption maxima of the oxovanadium(IV)-levofloxacin complex in pure solvents follow the order: DMF>MeOH>H_2O. The oxovanadium(IV)-levofloxacin complex has a 1:2 stoichiometry in all pure and binary mixed solvents. The logβ value of the $[V^{IV}O(lev)_2]$ complex follows the order MeOH>H_2O>DMF. The composition of binary mixed solvents affects the logβ values of the $[V^{IV}O(lev)_2]$ complex. For a given binary mixture composition, the logβ value of the $[V^{IV}O(lev)_2]$ complex is greater in the MeOH-H_2O system than in the DMF-H_2O system. The logβ value decreases as the ionic strength and solution temperature increase. Thermodynamic parameters indicate that the formation of the oxovanadium(IV)-levofloxacin complex is spontaneous and exothermic.

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Authors contribution

Iffat Ara Shaon: Data curation, Formal analysis, Conceptualization; Subarna Karmaker: Writing–original draft, Data curation, Formal analysis, Funding acquisition, Conceptualization; Tapan Kumar Saha: Data curation, Formal analysis, Conceptualization, Funding acquisition, Resources, Writing – review and editing, Investigation, Supervision.

Conflict of interest

The authors declare no competing financial interest.

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