



Synthesis, Characterization and Biological Applications of Mo(VI) and V(IV) Complexes of 2-[2-(2-aminoethoxy)ethoxy]ethanamine dithiocarbamate

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ABSTRACT

New dithiocarbamate(DTC) 2-[2-(2-aminoethoxy)ethoxy]ethanamine dithiocarbamate (AEEEEA DTC) ligand (L) was synthesized by the reaction of diamine and carbon disulfide with 1:2 molar ratio. Molybdenum (VI) and vanadium (IV) complexes of this ligand, ([MoO₂L] and [VOL]) were prepared by 1:1 molar proportion. Prepared ligand and complexes were characterized using molar conductance, elemental analysis, UV- visible, FT-IR and ¹H-NMR spectroscopy. In the FT-IR spectra, band for S-H of the ligand disappeared in the complex confirming the sulphur-metal(S-M) linkage. The presence of peak for S-H proton in the ¹H-NMR spectrum of ligand and its disappearance in the complex supported the FT-IR result. Low molar conductivity indicated the non-electrolytic nature of the complexes. The electronic spectra and effective magnetic moment of the prepared complexes revealed the +4 and +6 oxidation state of vanadium and molybdenum, respectively. Antimicrobial activities were investigated for the synthesized ligand.

Keywords: 2-[2-(2-aminoethoxy)ethoxy]ethanamine dithiocarbamate (AEEEEA DTC), Mo(VI) and V(IV) complexes of AEEEEA DTC, FT-IR, ¹H-NMR, antibacterial and antifungal activity.

1. INTRODUCTION

Dithiocarbamates (DTC) are well-known compounds for binding strongly and selectively to many metals. Therefore, in the past few decades self-assembly directed by metal–dithiocarbamate coordination has emerged as a useful supramolecular methodology for the preparation of macrocycles, cages, catenanes and nanoparticles [1]. The strong metal binding property of dithiocarbamic acid and their chelates play an important role in biological, agricultural and industrial fields [2]. The

chelating property of DTC arises from a planar delocalized π -electron flow from the nitrogen to the sulphur [1]. The net effect of electron flow facilitates strong donation from ligand to metal, resulting in high electron density on the metal sphere and stabilizes high oxidation states in metal complexes [3]. Therefore, dithiocarbamates are effective ligand with binding flexibility and have a variety of applications. The extensive biological applications of dithiocarbamate complexes, such

as anti-bacterial, anti-tumor, anti-tubercular [4, 5], etc. have already been reported. The tailoring of new dithiocarbamate attracts research interest to new applications. This area is still in its early stages but some applications have already been noted including the functionalization of gold nanoparticles, stepwise build-up of multimetallic arrays, dithiocarbamate-containing supramolecular systems for anion binding, development of technetium radiopharmaceuticals [6]. Begum *et al.* have synthesized mixed ligand catecholato-bis(diamine-mono-dithiocarbamate) vanadium(IV) complexes using ethane-1,2-diamine, propane-1,3-diamine, *N,N*²-diethylethane-1,2-diamine and hexane-1,6-diamine [7]. Chowdhury *et al.* have synthesized dioxo-molybdenum(VI) complexes of mono-dithiocarbamate using ethane-1,2-diamine, propane-1,3-diamine, *N,N*-dimethylethane-1,2-diamine, *N,N*-diethylethane-1,2-diamine, hexane-1,6-diamine and they observed the bidentate nature of ligand [8]. Halimehjani *et al.* have prepared dithiocarbamates of *N',N''*-dicinnamylalkane-1,*n*-diamines with Zn, Ni, Pd, Hg and Pb and noticed the amorphous phase of the complexes [9]. But the study on designing of dithiocarbamate by using ether type diamine viz. 2-[2-(2-aminoethoxy)ethoxy]ethanamine (AEEEE) has not yet been done. AEEEE is commonly known as 2,2'-(ethylenedioxy)bis(ethylamine) and is used in the production of many industrial chemicals. Karadağ *et al.* have shown that 2,2'-(ethylenedioxy)bis(ethylamine) can coordinate tridentately with Ni(II) and tetradentately with Cd(II) from their saccharinates. It revealed the tridentate and μ -N/O bidentate bridging behavior of Cu(II) complexes [10]. Şenocak *et al.* have used 2,2'-(ethylenedioxy)bis(ethylamine) to synthesize cyano-bridged ligand and their bimetallic Ni, Cu complexes [11]. Devi *et al.* have synthesized the Schiff base ligand of 5-chlorosalicylaldehyde by reacting with 2,2'-(ethylenedioxy)bis(ethylamine) and 1-hydroxy-2-naphthaldehyde/2-hydroxy benzaldehyde. Later they employed the ligands to make organosilicon and organotin complexes [12, 13]. They also screened

the antibacterial activity of the complexes. A new imine-ether ligand *N,N*-bis(2-pyridylmethylene)-2,2'-(ethylenedioxy)bis(ethylamine) has been synthesized by Vallina *et al.* and they studied their coordinating property [14]. Macrocyclic Schiff bases of 2,2'-(ethylenedioxy)bis(ethylamine) with 1,7-bis(6-methoxy-2-formylphenyl)-1,7-dioxahseptane have been studied by Kedy *et al.*, and observed good Ag⁺ extracting power from aqueous to organic phase [15]. Leon *et al.* have prepared poly [*N*-iso propyl acrylamide-2,2'-(ethylenedioxy)bis(ethylamine)-2,3,5-triiodobenzamide] and reported their radio-opaque thermosensitivity [16]. However, to the best of our knowledge, no published work is available on the preparation of dithiocarbamate by using AEEEE. The AEEEE possesses the linking capability for conjugation [16]. Consequently, the preparation of new dithiocarbamate ligand using this ether type diamine and its complex of bioactive metals like molybdenum and vanadium may compete for interesting applications. Therefore, in this research, an attempt has been taken to synthesize AEEEE DTC (L) ligand and its molybdenum(VI) and vanadium(IV) complexes as a new approach. The synthesized ligand and complexes are characterized and then screened for biological application.

2. MATERIALS AND METHODS

2-[2-(2-aminoethoxy)ethoxy]ethanamine (AEEEE) was purchased from Sigma Aldrich to prepare dithiocarbamate ligand with CS₂ (Scharlau, Spain). Sodium molybdate dihydrate (BDH, UK) and vanadyl sulfate (E. Merck, Germany) were used in synthesizing complexes. Methanol, ethanol, and DMSO (Scharlau, Spain) and DMF from Guangdong, China were used as supplied. All of the chemicals and solvents were used without further purification.

2.1 Preparation of AEEEE DTC Ligand (L)

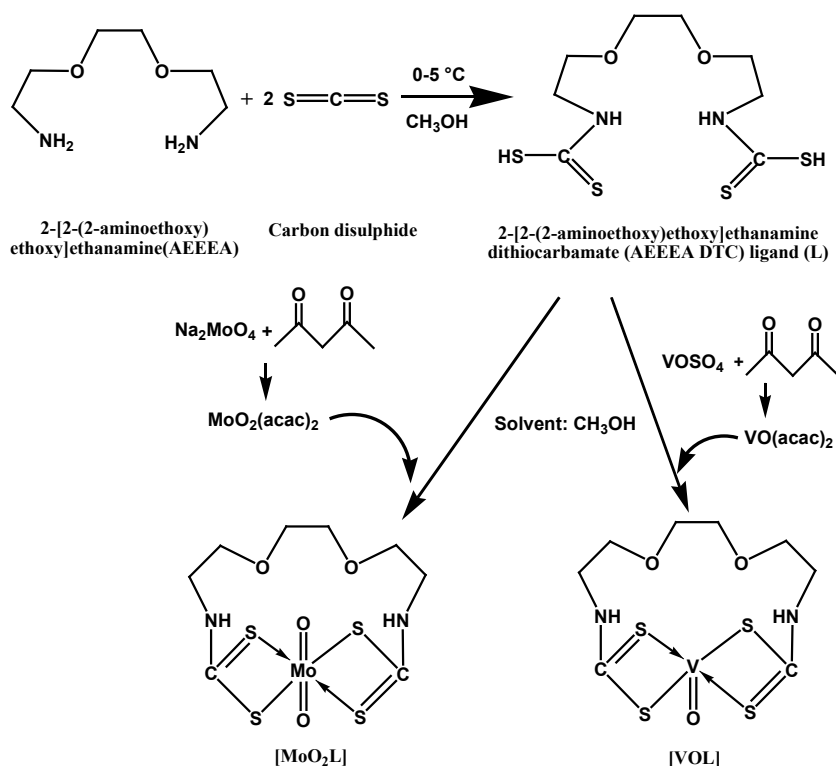
AEEEE DTC (L) was prepared by the reaction of diamines with carbon disulfide at 1:2 molar proportions. To prepare the desired

dithiocarbamate ligand, 50 mL methanolic solution of AEEEA (4 mmol) diamine was allowed to cool in a freezing mixture of ice and salt bath (0-5 °C). To this, carbon disulfide (8 mmol) was added drop-wise for about half an hour with constant stirring. After 24 hours white precipitate was formed. The precipitate was allowed to stand in the ice-salt bath and thereafter filtered off, washed with methanol and dried over silica gel in a vacuum desiccator. The yield was 70% and melting point was 110 °C. The possible chemical reaction is shown in **scheme 1**.

2.2 Preparation of Metal Acetylacetonate

Bis(acetylacetonato)dioxomolybdenum(VI), $\text{MoO}_2(\text{acac})_2$ and bis(acetylacetonato)oxovanadium(IV), $\text{VO}(\text{acac})_2$ were synthesized as intermediate for the synthesis of Mo(VI) and V(IV) complex,

respectively [7, 8]. Sodium molybdate dihydrate (0.494 g; 2.04 mmol) was dissolved in water (50 mL). Hydrochloric acid (6 N) was added to this solution until it reached pH=1. Then acetylacetonate (0.62 mL; 6 mmol) was added to this solution with vigorous stirring to break up globules which contained entrapped acetylacetonate. The greenish-yellow solid formed was isolated by filtration, washed with cold ethanol and acetone and dried over silica gel in a vacuum desiccator. To prepare bis-(acetylacetonato)oxovanadium(IV), 0.62mL acetylacetonate was added drop-wise with a 50 mL aqueous solution of vanadyl sulfate (0.332 g) with stirring. Anhydrous sodium carbonate was added to neutralize the solution. The mixture was cooled in an ice salt bath and the resulting precipitate was filtered, washed with cold distilled water, and dried in a vacuum desiccator.



Scheme 1: Possible reaction pathway of the synthesis of ligand (L) and its $[\text{MoO}_2\text{L}]$ and $[\text{VOL}]$ complexes.

2.3 Preparation of Molybdenum (VI) and Vanadium (IV) Complexes

The molybdenum (VI) [MoO₂L] and vanadium (IV) [VOL] complexes of AEEEE DTC ligand (L) were prepared by the condensation reaction of metal acetylacetonate {[MoO₂(acac)₂] and [VO(acac)₂]} and ligand (L) in 1:1 molar proportion. 1 mmol (0.301 g) of prepared dithiocarbamate was added to a 50 mL methanolic solution of metal acetylacetonate {1 mmol, 0.326 g of [MoO₂(acac)₂] and 1 mmol, 0.265 g of [VO(acac)₂]}. With the addition of ligand, the precipitate came out almost immediately. After 24 hours the color of the precipitate became intensify. The precipitate was filtered off, washed with methanol and dried over silica gel in vacuum desiccators overnight. Molybdenum (VI) complex [MoO₂L] appeared dark green color with 70% yield and the melting point 220 °C. Vanadium (IV) complex [VOL] looked green in color with 72% yield and the melting point 220 °C. The possible chemical reactions for the preparation of [MoO₂L] and [VOL] complexes are shown in **scheme 1**.

2.4 Analysis

Melting points of the ligand and complexes were obtained with Melting Point Apparatus, model KRUSS, KSPINN. The elemental analysis of the sample was performed by Elementar (Vario-EL cube) and metal analysis by Shimadzu

AA-7000, AAS using GFA-7000. CTS-406K model conductometer was used for the measurement of conductance of the samples in DMF and DMSO solvent. The FT-IR spectra were recorded by FT-IR 8400S Shimadzu spectrophotometer in the range 4000- 400 cm⁻¹ at 2 cm⁻¹ resolution and 30 times scanning using KBr. ¹H-NMR spectra were recorded by NMR spectrometer; model Ascend-400, BRUKER. DMSO-*d*₆ was used as a solvent in the ¹H-NMR analysis. The chemical shift of DMSO-*d*₆ is 2.49 ppm [17]. The electronic absorption spectroscopic study of the prepared samples in DMSO and DMF was done by running on a Shimadzu UV-Visible spectrophotometer (model-UV 1800) using 1 cm cell. Physical data of the prepared ligand and complexes are shown in **Table 1**.

2.5 Biological Activity Test

The biological activity was done in the disc diffusion method [18]. Mueller-Hinton Agar was used as nutrient media for the microorganism. In a sterile petri dish, the sterilized neutral media (autoclaving at 121 °C and 15 lbs/sq. inch pressure for 20 minutes; at 25 °C maintaining pH at 7.2 ± 0.2) was incubated at 30-37 °C for 30 minutes. The pure cultured microbes were inoculated to Trypticase soy broth and incubated at 37°C until achieving the turbidity of the 0.5 McFarland standard and then it was dispersed on agar in a

Table 1. Physical data and effective magnetic moment of the ligand and its complexes of molybdenum (VI) and vanadium (IV).

Ligands/ Complexes	Elemental Analysis						Molar Conductance (Λ _m) (μS)	μ _{eff} (B.M.)
	C% (cal)	H% (cal)	N% (cal)	S% (cal)	Mo% (cal)	V% (cal)		
L	32.41 (31.98)	5.62 (5.37)	8.96 (9.32)	40.11 (42.68)			0.01	
[MoO ₂ L]	22.07 (22.53)	3.60 (3.31)	6.15 (6.57)	28.27 (30.08)	21.15 (22.50)		0.02	0
[VOL]	25.73 (26.30)	4.21 (3.86)	7.24 (7.67)	32.71 (35.10)		13.24 (13.94)	0.01	1.37

petri dish. In the meantime solutions of known concentration ($\mu\text{g mL}^{-1}$) of the test samples were prepared in DMSO. Discs containing the test material were placed on dry inoculated nutrient agar medium uniformly seeded with the test microorganisms. Standard antibiotic discs and blank discs (impregnated with solvents) were used as positive and negative control. The plates were incubated at 37°C and inverted within 15 minutes after the discs were applied. After 16 to 18 hours of incubation the zones of inhibition produced by compounds and standards were recorded in mm and compared. The antibacterial activity of synthesized ligand was studied against some gram-positive bacterial strains like *Bacillus cereus*, *Bacillus subtilis*, *Staphylococcus aureus* and gram-negative strain *Shigella dysenteriae*. Ciprofloxacin was used as a standard antibiotic in the antibacterial test. *Candida albicans* and *Aspergillus niger* fungi were used for the test of antifungal activity of ligand using Fluconazole as standard. The antifungal activity test was also done by the disc diffusion method.

3. RESULTS AND DISCUSSION

3.1 Elemental (C, H, N, S) Analysis

Elemental data of the prepared AEEEEA DTC ligand (L) and its molybdenum (VI) and vanadium (IV) complexes ($[\text{MoO}_2\text{L}]$, $[\text{VOL}]$) are shown in **Table 1**. The analytical data for C, H, N, S elements of both ligand and complexes are almost similar to the calculated values. It indicates that the synthesized ligand and complexes may have almost similar elemental composition to the expected structures.

3.2 Molar Conductance Measurement

Conductance is an expression with which electric current flows through a substance. 10^{-3} M solution of ligand and complexes in DMSO and DMF was used for conductivity measurement at 30°C . Experimental molar conductance (Λ_m) values are shown in **Table 1**. Low molar conductivity values are observed for all of the complexes which indicate their non-electrolytic nature [19, 20]. It reveals that the ligand is covalently bonded to the central metal.

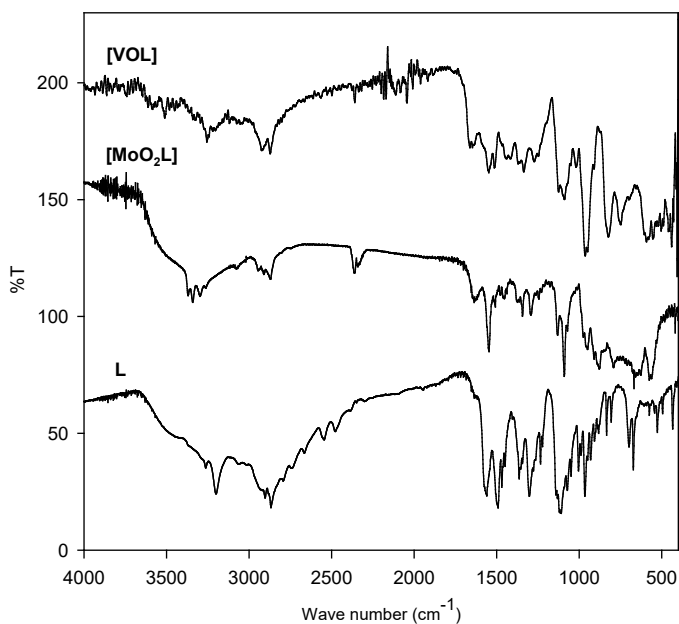


Figure 1. FT-IR spectra of synthesized ligand (L), $[\text{MoO}_2\text{L}]$ and $[\text{VOL}]$ complexes.

3.3 FT-IR Spectra

The infrared studies of metal complexes help to illustrate the coordination of the ligand and assume structural properties. FT-IR spectra of the ligands and complexes are shown in Figure 1. The main vibrational modes in dithiocarbamate ligand are C-N, C-S, CSS and S-H. It was reported that, IR band at 1350-1450 cm^{-1} region associated with C-N; 950-1080 cm^{-1} for CSS; 2440-2580 cm^{-1} for S-H [20, 21, 22]. IR spectra at 1104-1200 cm^{-1} and 760 cm^{-1} were assigned for C=S (thio keto) and C-S(thiolato) vibration, respectively [23]. The band in the region of 3150-3300 cm^{-1} was recognized for the N-H vibrational mode [8, 24]. FT-IR spectrum of AEEEEA DTC (L) contains bands C-N at 1382 cm^{-1} along with C=S at 1112 cm^{-1} and C-S at 700 cm^{-1} stretching modes. These suggest the attachment of CS_2 with diamine and support the formation of the proposed ligand. The presence of bands C-N, C=S and C-S in the molybdenum and vanadium complexes refer to the existence of ligand in the complexes. The FT-IR spectrum of ligand possesses characteristic band for S-H at 2544 cm^{-1} which gets disappeared in the complex. This implies that both the sulphur atoms of the ligand take part in coordination through the deprotonation of thiol (S-H). The shifting of the CSS single band to the higher wavenumber in the complex also indicates the coordination of ligand. The existence of N-H stretching band in both ligand and complexes signifies that the nitrogen of N-H did not take part in the coordination with the central metal. M=O and M-S regions (where M indicates the metal Mo and V) are characteristic of dithiocarbamate complexes. Bands for M-S (thiol) and M-S (thio keto) stretching vibrations were observed at 617-670 cm^{-1} and 400-420 cm^{-1} regions, respectively [8]. Thus the bands appeared at 418 and 460 cm^{-1} for thio keto and 685 and 610 cm^{-1} for thiolato form in complexes of $[\text{MoO}_2\text{L}]$ and $[\text{VOL}]$, respectively reveal that the central metal attached with the sulphur of the ligand. It was reported that, at 910-991 cm^{-1} region, two symmetric O=Mo=O

stretching bands indicated the cis- MoO_2^{2+} form whereas a single band for asymmetric O=Mo=O revealed trans- MoO_2^{2+} structures [25, 26]. Since $[\text{MoO}_2\text{L}]$ comprises an FT-IR band at 950 cm^{-1} indicates the trans- MoO_2^{2+} moiety in the proposed complex. The band at 962 cm^{-1} attribute to the stretching vibration of the terminal V=O bond in the $[\text{VOL}]$ complex [27].

3.4 Electronic Spectra

Ultraviolet spectroscopy is a useful tool in the characterization of organometallic compounds. In the visible region, the electronic transitions in the crystal field depend largely on the metal ion. The UV-Vis spectra of the samples were measured in DMSO and DMF as 10^{-5} M in the range 250-700 nm. The electronic spectra of the ligand and its complexes are shown in Figure 2.

For multiple bonds in the molecule, the $\pi \rightarrow \pi^*$ and $n \rightarrow \pi^*$ transitions occur and hence these phenomena increase considerably in wavelength and intensity. The peaks at 250-350 nm were assigned to the intra-molecular intra-ligand transitions corresponding to $\pi \rightarrow \pi^*$ and $n \rightarrow \pi^*$ of the N-C=S and S-C=S group [28]. UV peak observed at 268 nm region in the spectrum of $[\text{MoO}_2\text{L}]$ complex. Beyond 500 nm, optical density values were extremely low indicated the absence of d-d transitions over the visible region [28]. Thus the electronic spectral data revealed the d^0 system of central metal molybdenum which confirms its genuine +6 oxidation state in the complex [28]. The complex is however, colored only through absorptions tailing from the ultraviolet. The vanadium (IV) complex has a band at 260 nm corresponding to a $\pi \rightarrow \pi^*$ transition in the near-ultraviolet region [28]. For the d^1 (vanadium-IV) system, a single absorption band for ${}^2\Gamma_{2g} \rightarrow {}^2E_g$ transition might be expected at 500 nm, [8] but unfortunately, in the present complex, there was no absorption band for ${}^2\Gamma_{2g} \rightarrow {}^2E_g$ transition.

3.5 ${}^1\text{H-NMR}$ Analysis

${}^1\text{H-NMR}$ of the ligand (L) and its molybdenum

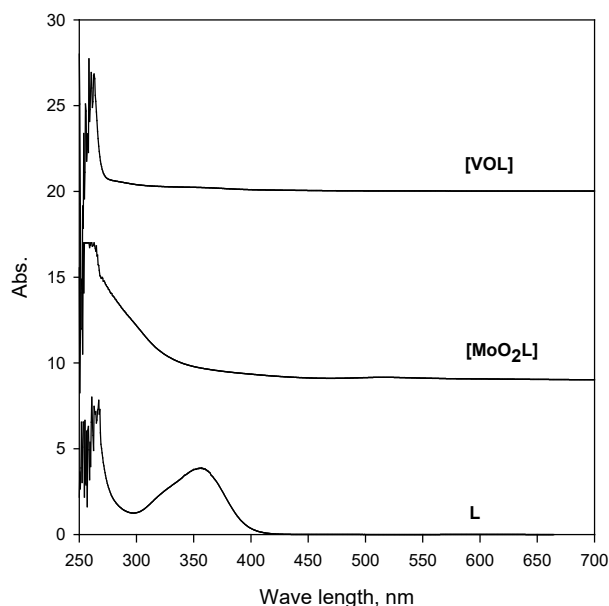


Figure 2. UV-vis spectra of synthesized ligand (L), [MoO₂L] and [VOL] complexes.

complex of [MoO₂L] were taken by dissolving in DMSO and are shown in Figures 3 and 4, respectively.

The ¹H NMR spectrum of ligand (400 MHz, DMSO-*d*₆) δ (ppm): 1.917 (s, 2H, *J* = 7.2 Hz), 2.961-3.000 (m, 4H, *J* = 5.2 Hz), 3.541-3.609 (t, 4H), 3.716-3.740 (t, 2H, *J* = 4.8 Hz), 7.523-7.889 (br, 2H).

In ¹H NMR spectrum of [MoO₂L] (400 MHz, DMSO-*d*₆) δ (ppm): 2.982-3.018 (q, 4H, *J* = 4.8 Hz), 3.507-3.555 (m, 4H, *J* = 6.4 Hz), 3.756-3.836 (m, 4H, *J* = 5.3 Hz), 7.538-7.989 (br, 2H).

Protons of CH₂ and S-H generally show chemical shift at 3.5-4.0 ppm [29] and 1.57-2.0 ppm [3, 30], respectively. The ¹H-NMR of the ligand contains the corresponding peak for one proton, in the region 1.92 ppm, ascribed to hydrogen linked directly with S atom in dithiocarbamate ligand and this peak gets disappeared in the spectrum of the complex [3, 30]. It suggests the coordination between ligand and metal occurred through the S moiety. A chemical shift δ = 2.96-3.84 ppm for CH₂ is observed in the prepared ligand and

complex [29]. According to the literature, the peak at 8.13 ppm in the ¹H-NMR spectrum corresponds to the proton of >N-H [7, 31]. This might be due to the same order o-donation and n-back donation characteristics in the sulphur atom of dithiocarbamate [3]. In the present work, the peak obtained in the range of δ=7.52-7.89 ppm for ligand and δ=7.54-7.99 ppm for the complex may be due to the NH proton [31, 32]. The downfield chemical shifting can be explained with the n(nonbonding) electron flow from nitrogen to sulphur causes the decrease of electron cloud on N along with the presence of electronegative oxygen atom on the aliphatic chain. The peak for NH proton in the ligand and complex indicates that there is no participation of the N-H site to the coordination with metal. The additional peaks in the NMR spectra revealed the presence of some impurities along with the desired product those were not identified. However, the presence of peak for the proton of the thiol group (S-H) in the ligand and its disappearance in the corresponding molybdenum complex support the existence

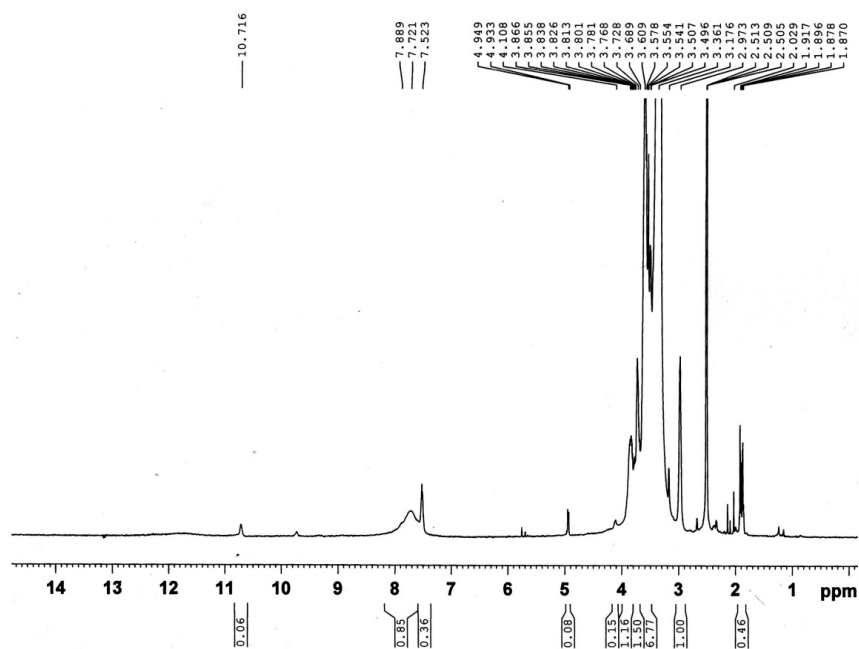


Figure 3. ^1H NMR spectrum of ligand (L).

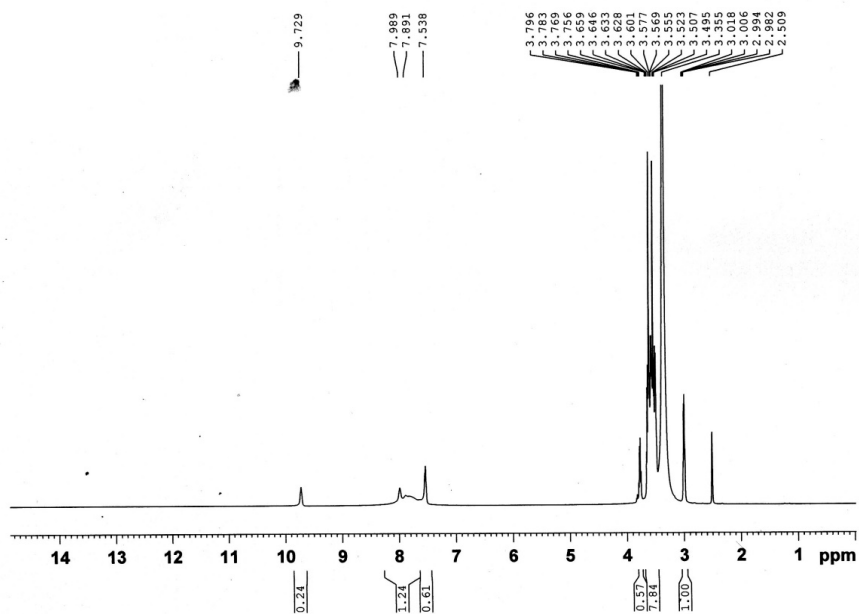


Figure 4. ^1H -NMR spectrum of molybdenum complex $[\text{MoO}_2\text{L}]$.

of AEEEA DTC ligand (L) and formation of $[\text{MoO}_2\text{L}]$ complex through SS coordination.

3.6 Magnetic Susceptibility Measurement

The magnetic susceptibilities of the prepared complexes were investigated using Gouy's method and the obtained data are presented in **Table 1**. The zero effective magnetic moment of $[\text{MoO}_2\text{L}]$ complex indicates the $4d^0$ electronic configuration of the molybdenum ion. Therefore, molybdenum possesses the +6 oxidation state in $[\text{MoO}_2\text{L}]$ complex. The vanadium complex ($[\text{VOL}]$) shows positive molar magnetic susceptibility that suggests its paramagnetic nature. The 1.37 BM of effective magnetic moment reveals the existence of one unpaired electron i.e. d^1 system in vanadium. This supports the presence of the +4 oxidation state of vanadium in $[\text{VOL}]$ complex.

3.7 Antibacterial Screening for Synthesized ligand

The antibacterial screening measures the ability of each antibacterial agent to inhibit the in vitro bacterial growth. The study of antibacterial activity of synthesized ligand was carried out

against some human pathogenic bacteria like *Bacillus cereus*[A], *Bacillus subtilis*[B], *Staphylococcus aureus*[C] and *Shigella dysenteriae*[D] by disc diffusion method using Ciprofloxacin as a standard [33, 34] maintaining pH at 7.2 ± 0.2 and shown in **Figure 5**. At 200 ppm concentration, the ligand shows the highest antibacterial activity against the *Bacillus cereus*. It also shows comparable bacterial inhibition against the remaining *Bacillus subtilis*, *Staphylococcus aureus* and *Shigella dysenteriae* at 200 ppm. At lower concentrations e.g. 10, 25, 50, 100 ppm the sample didn't give any remarkable inhibition against the bacteria. This may be due to the higher growth rate of bacteria than the available dose of diffused samples capable to alter the metabolic activity in the bacterial cell.

3.8 Antifungal Screening for Synthesized Ligand

Candida albicans and *Aspergillus niger* fungi were screened for antifungal activity of ligand using Fluconazole as standard. Antifungal activity test was investigated by the disc diffusion method at the concentrations of 25, 50, 200, 500 and 1000 ppm in DMSO. There was no notable inhibition against the tested fungi.

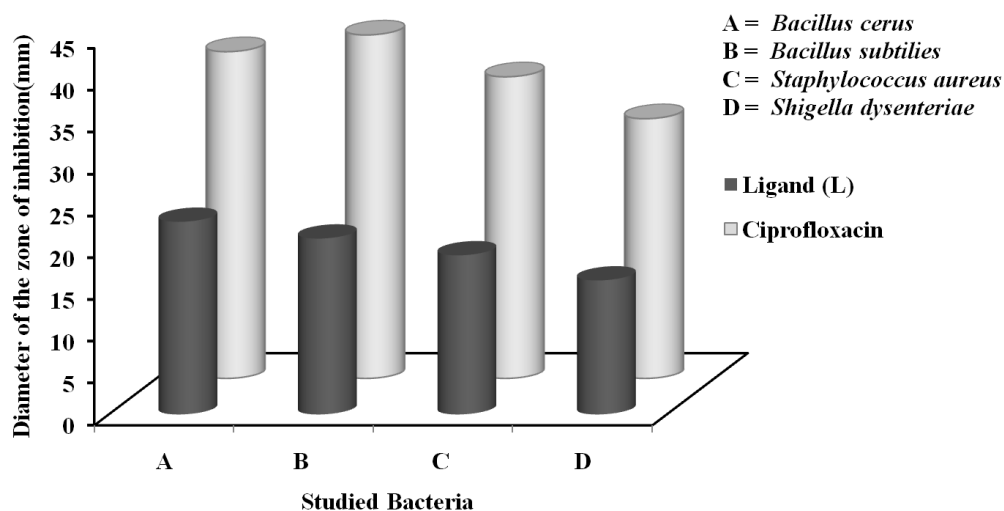


Figure 5. Inhibition zones of the ligand (L) at 200 ppm against bacteria

4. CONCLUSIONS

Novel AEEEA DTC ligand (L) was synthesized by using ethereal diamine AEEEA. The ligand was then used to prepare molybdenum and vanadium complexes ($[\text{MoO}_2\text{L}]$ and $[\text{VOL}]$). The FT-IR spectra of the synthesized ligand showed all characteristic bands and suggested the formation of complexes through the S-S donor site. The $^1\text{H-NMR}$ also supported the coordination of ligand to the metal. Magnetic susceptibility data demonstrated the diamagnetic nature of $[\text{MoO}_2\text{L}]$ and revealed the +6 oxidation state of molybdenum which was also supported by UV data. The d^1 state of vanadium was observed from the paramagnetic value for $[\text{VOL}]$. From the overall analysis, octahedral geometry of molybdenum (VI) complex and the square pyramidal geometry of vanadium (IV) complex of AEEEA DTC might be proposed. It is required for more analytical support such as a crystallographic study to confirm the actual structure of the synthesized complexes. In the microbial activity test, the ligand exhibited antibacterial behavior against all of the tested bacteria and inhibited *Bacillus cereus* more effectively than other experimented microbial strain.

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