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Design, Synthesis, Structural Elucidation And Dna Cleavage Studies Of Acenaphthenequinone Derived Schiff Base Nickel (Ii) Complexes

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Abstract: The broad spectrum application of Acenaphthenequinone and its derivatives as biologically active compounds has turned the attention of the Chemists to review their chemistry. Three novalhexadentate Schiff base ligands have been synthesised by the condensation of Acenaphthenequinone with essential amino acids in 1:2 stoichiometry. Synthesized ligands after complexation with metal ionsare structurally characterized by analytical and spectral techniques. Thecomplexes are found to be non-electrolytic in nature.Antimicrobial activity of the metal complexes was tested using agar well diffusion method against the gram positive and negative strains and found to be potent biocidal agents. Themetal complexes are subjected to DNA cleavage with pUC19DNA.

Key Words: acenaphthenequinone, aminoacids, DNA Cleavage, antibacterial activity.

1. Introduction

Amino acids constitute the building blocks of proteins and are indispensible for performing large number of biological functions [1,2].The mononuclear and multinuclear metal complexes have great impact in different areas of chemistry like organic, inorganic, bio-inorganic and medicinal chemistry.Acenaphthenequinone derivatives are found to be useful intermediate in the manufacture of dyes.[3-5]Schiff bases derived from acenaphthenequinone have potent applications in biology as bactericidal, antihypoxic, and fungicidal agents and are useful as phospholipase A2 inhibitors[6]. Multiple functional groups (COOH, NH_2) present in the amino acid side chain of the ligands can lead to the

ISSN:0971-1260
Vol-22-Issue-4-October-December-2019

development of unexpected and unusual structures [7,8]. Here we have reported the synthesis, characterization and DNA cleavage studies of the nickel complexes of Acenaphthenequinone with essential amino acids like glycine, cysteine and methionine.

2.Experimental

2.1 Materials

All the reagents involved in this work were purchased in pure form and used as such. Solvents used were either of 99% purity or purified by known laboratory procedure [9]. TrisHCl buffer, Nutrient medium were purchased from Himedia. pUC 19 DNA was procured from Genie Company, Bangalore.

2.2 Instrumental

The elemental analysis of Carbon, Hydrogen and Nitrogen was recorded on Elementer Vario EL III elemental analyser. The mass spectral analysis was carried out in JEOL-300 mass spectrometer. The room temperature FT-IR spectra for the ligand and complexes were recorded as KBr pellets with SHIMADZU FT-IR 8400S spectrometer in the range of 4000-400cm^{-1}. The molar conductance values for the complexes were measured using Systronics Conductivity bridge type using DMF solvent. UV-Visible spectra were recorded using Perkin Elmer Lambda 35 UV-Visible spectrophotometer in the range of 190-1100 nm using DMF as the solvent. The X-band ESR spectrum of the copper complexes in DMSO was recorded with JEOL model JES FA200 spectrometer at Liquid Nitrogen temperature (LNT).Cyclic Voltametric studies of the synthesized complexes were carried out at room temperature with Impedenceanalyzer cum cyclic voltameter, VersaSTAT MC in DMF solution containing 0.1M $NaClO_4$using a glassy carbon electrode.

2.3 Synthesis of Schiff base ligand and its metal complexes

2.3.1 Synthesis of Schiff base ligand

ISSN:0971-1260
Vol-22-Issue-4-October-December-2019

2 mmol of Acenaphthenequinone (A) and 4 mmol of the amino acid (L-Glycine(B)/L-Cysteine(C)/ L-Methionine(D) were dissolved in 40 ml of warm Dichloromethane. The reaction mixture was stirred overnight on a magnetic stirrer to give yellow coloured precipitate. The precipitate was washed with water and then with diethylether. It was then dried in vacuum.

2.3.2 Synthesis of Schiff base metal complexes

An equimolar (1mmol) methanolic solution of the Schiff base ligand (AB/AC/AD) and Nickel(II)chloride hexahydrate were stirred and refluxed at 50°C for 8 hours. The precipitated metal complexes were filtered, washed with ethanol, ether and dried in vacuum.

2.4 DNA Cleavage

The DNA Cleavage studies of the synthesized molecules were carried out by agarose gel electrophoresis method [10]. The gel electrophoresis experiments were performed by the incubation of the samples containing 40μm pUC DNA, 50μm metal complexes and 50μm H_2O_2 in Tris-HCl buffer (7.7) at 37°C for 2 hours. After incubation, the samples were subjected to electrophoresis for 2 hours at 50V on 1% agarose gel using Tris-acetic acid-EDTA buffer. The gel was then stained using 1μg cm^{-3}ethidium bromide (EB) and photographed under ultra violet light at 360nm [11].

2.5 Antimicrobial Studies

The antibacterial efficacy of the synthesized complexes was tested against *Bacillus subtilis, Staphylococcus aureus, Escherichia coli, Klebsiella pneumonia* and *Pseudomonas aeruginosa* by agarwell diffusion method. Muller-Hinton agar (MHA) plates were swabbed with overnight culture of respective bacteria. Wells were made in each of these plates using sterile cork borer. Required amount of the solution of metal complexes were added into the wells and allowed to diffuse at room temperature for 2hrs. Control

ISSN:0971-1260
Vol-22-Issue-4-October-December-2019

experiments comprising inoculums with cefatoxime disc were set up. The plates were incubated at 37°C for 18-24 h. The diameter of the inhibition zone (mm) was measured and the activity was calculated.

3.Results and Discussion

All the synthesized complexes are insoluble in common organic solvents but are soluble in DMSO and DMF.

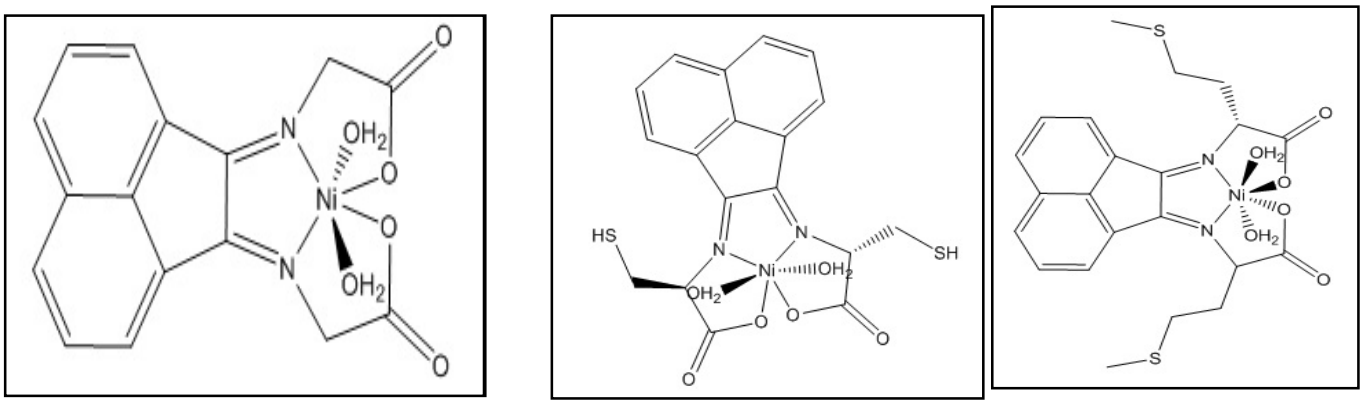
3.1 Elemental Analysis and Molar conductance

The mononuclear NiAB/NiAC/NiAD complex is stable towards air and is non-hygroscopic in nature.The microanalysis for carbon, hydrogen and nitrogen and the physical data of the synthesized complexes are given in Table 1.This experimental data corresponds to the suggested molecular formula of the complexNiAD, viz.$C_{22}H_{24}N_2O_6S_2$ Ni.The structure of the synthesized complexes NiAB, NiAC, NiAD are given in Fig.1.

Table1. Analytical Data of the Schiff base complexes

Compound	Empirical Formula	Yield (%)	% of elements, Found (Calc)				Molar conductance	Magnetic moment (BM)
			C	H	N	Cu		
NiAB	$C_{16}H_{14}N_2O_6$.Ni	67	49.40 (49.42)	3.6 (3.9)	7.2 (7.1)	15.11 (15.09)	23.02	2.98
NiAC	$C_{18}H_{18}N_2O_4S_2$. Ni	64	66.59 (66.54)	4.91 (4.87)	8.47 (8.44)	14.19 (14.15)	14.9	3.38
NiAD	$C_{22}H_{24}N_2O_6S_2$. Ni	63	52.38 (52.43)	3.91 (3.97)	5.48 (5.56)	12.59 (12.62)	24.1	1.84

ISSN:0971-1260
Vol-22-Issue-4-October-December-2019



(a) (b) **(c) Fig.1.Structure of (a) NiAB (b) NiAC (c) NiAD**

3.2 ^{13}C NMR Spectral Studies

The ^{13}C NMR spectrum of the Schiff base ligand AD,AC and AB recorded in $CDCl_3$ is given in Fig.2, S1, S2. It exhibits 4 signals confirming the presence of 4 different types of carbon atoms. The set of signals at 122.24 to 144.29 ppm is assigned to aromatic carbon atoms, Carboxylic carbon is observed at 187.5 ppm and the signal at 163.18 ppm confirms the presence of azomethine carbon, thus authenticating the formation of Schiff base.

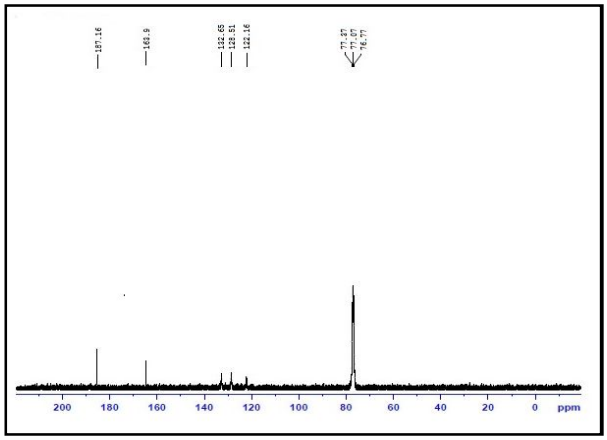


Fig.2. ^{13}C NMR spectrum of the Schiff base ligand AD, recorded in $CDCl_3$

3.3 Fourier Transform-Infrared (FT-IR) spectra

ISSN:0971-1260
Vol-22-Issue-4-October-December-2019

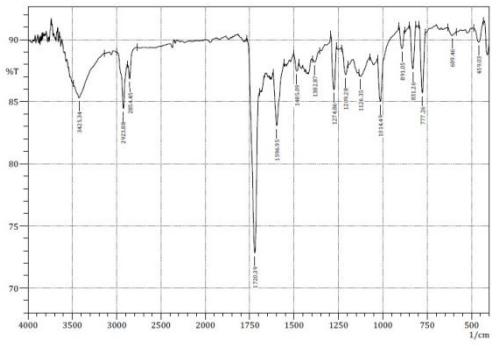


Fig.3.Vibrational Spectrum of Schiff base (AD)

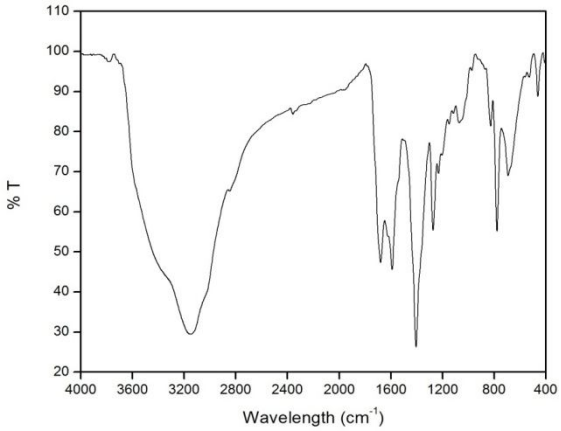


Fig.4. Vibrational Spectrum of NiAD

Table.2. IR Spectral data of Schiff base and its nickel complexes

Compound	ν(C=N)	$\nu(COO^-)_s$	$\nu(COO)_{as}$	ν(M–O)	ν(M–N)
AB	1598	1449	1718	---	---
AC	1596	1722	1484	---	---
AD	1596	1720	1485	---	---
NiAB	1586	1702	1424	421	528
NiAC	1589	1677	1406	459	530
NiAD	1588	1679	1405	459	516

FTIR spectrum of the NiAD complex is depicted in **Fig 4** and the spectral data in **Table 2**. The spectrum explains that imine (C=N) group has been shifted from 1596 to 1588 cm^{-1}confirming the co-ordination of the free ligand with the central metal ion through imine nitrogen [12]. The asymmetric stretching frequency due to the carboxylic acid in the free ligand at 1720cm^{1}has been shifted to 1679 $cm^{-1}$and that due to symmetric stretching at 1485 cm^{-1}has been shifted 1405cm^{-1}confirming the bonding of the Ni(II) ion by the carboxylato oxygen atom. Also additional bands in the region 459 cm^{-1}, 516 cm^{-1} confirms the formation of M–O and M–N bonds. The broad band at 3186cm^{-1} is due to intermolecular hydrogen bonding of –OH stretching resulting from water molecules present inside the coordination sphere of the complex. Thus in NiAD, Schiff base AD is coordinated to Ni(II) ion in tetradentate manner, through two imino nitrogen atoms and two carboxylato oxygen atoms of methionine moiety.

ISSN:0971-1260
Vol-22-Issue-4-October-December-2019

3.4 Electronic Spectroscopy

In NiAC complex [Fig.5]the first band appearing at 975nm corresponds to $^3A_{2g}(F) \rightarrow {}^3T_{2g}(F)$ transition. The second and third bands arising due to $^3A_{2g}(F) \rightarrow {}^3T_{1g}(F)$ and $^3A_{2g}(F) \rightarrow {}^3T_{1g}(P)$ are observed at 567 nm and 453 nm confirming octahedral geometry of nickel complex [7].

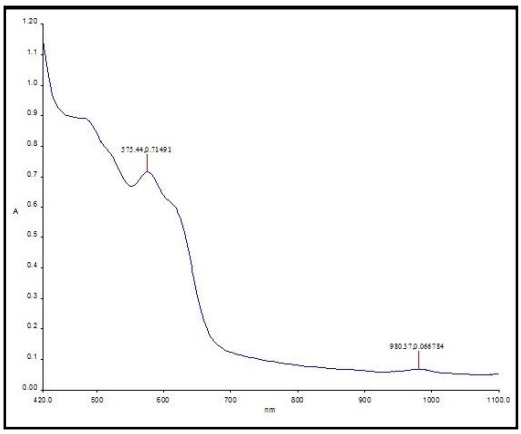


Fig. 5a. Electronic spectrum of NiAC complex (200-1100nm)

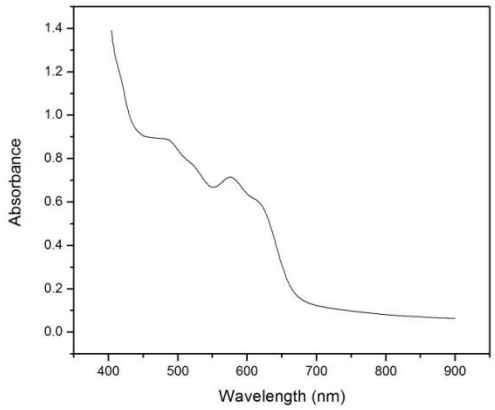


Fig. 5b. Electronic spectrum of NiAC complex (400-900nm)

3.5 Cyclic Voltammetry

The electro analytical technique is the most efficient and versatile method available for the mechanistic study of redox system. The Cyclic Voltammogram of NiAD complex, obtained in DMF solution at a scan rate of $0.5Vs^{-1}$ over the potential range of -1.5 to +2 V is given in Fig.6. The result exposes two redox stages. In the first, anodic peak (Ep_a) appeared at +1.4V and the cathodic peak (Ep_c) at +0.920V. This reveals an one electron quasi reversible (ΔEp=0.35V) process corresponding to Ni(I)/Ni(II) couple. In the next step, the anodic peak occurs at -0.75V and that of cathodic peak at -0.920V, analogous to one electron quasi reversible process of Ni(II)/Ni(III) couple [13].

ISSN:0971-1260
Vol-22-Issue-4-October-December-2019

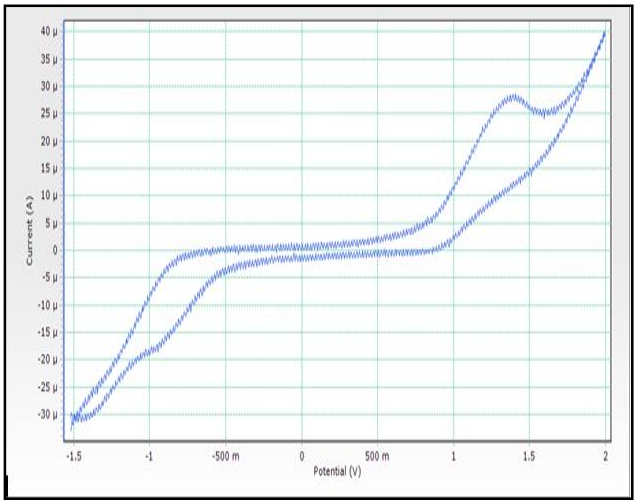
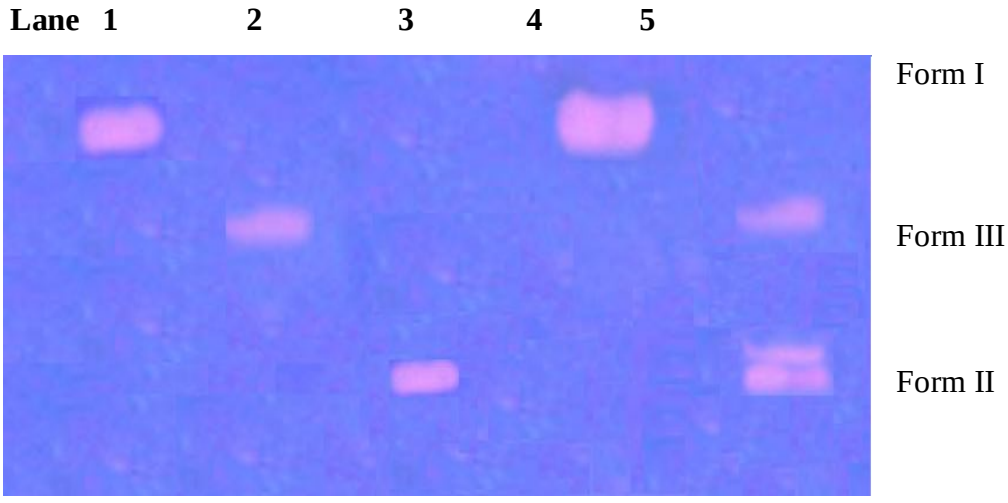


Fig.6. Cyclic Voltammogram of NiAC complex

3.6. DNA Cleavage Activity

Schiff base metal complexes when interact with DNA could induce the breakage of DNA by gel electrophoresis technique **[Fig.7]**. During DNA cleavage, the double strand breaks and the replication of the gene is thereby destroyed. In the present DNA cleavage study of Nickel complexes, Lane 1 (DNA control) and Lane 4 (NiAB) does not exhibit any cleavage. In Lane 3, the NiAC complex in the presence of H_2O_2 cleaves the DNA from supercoiled form (Form I) to nicked circular form (Form II). NiAD (Lane 5) complexes in H_2O_2 cleave the DNA from supercoiled form (Form I) to linear form (Form III). Thus, except NiAB all the other Nickel complexes are potent DNA cleaving agents.



ISSN:0971-1260
Vol-22-Issue-4-October-December-2019

Fig.7 Gel electrophoresis diagram showing the cleavage of pUC 19 DNA by the synthesized complexes in the presence of H_2O_2. Lane 1, DNA control;Lane 3, DNA + NiAC + H_2O_2; Lane 4, DNA + NiAB+ H_2O_2 and Lane 5, DNA + NiAD+ H_2O_2

3.8 Antimicrobial Screening

The antimicrobial data displayed in Table3 and Fig.8 clearly explains the activity of the synthesized metal complexes on selected gram negative bacterial strains like *Escherichia coli*, *Klebsiella pneumonia*, *Pseudomonas aeruginosa* and gram positive bacterial strains like *Bacillus subtilis*, *Staphylococcus aureus* by agar well diffusion method. All the complexes are active towards *Bacillus subtilis* and *Staphylococcus aureus*.NiAC complex shows no activity towards *Klebsiella pneumonia* and NiAD is passive towards *Pseudomonas aeruginosa*.The antimicrobial efficacy of the metal complexes is due to co-ordination and chelation [14,15]. As a result of chelation, the polarity of the central metal atom is reduced, because of the partial sharing of its positive charge with the ligand.

Table.3. Antimicrobial screening of nickel complexes by agar well diffusion method (zone formation in mm)

Compound	*Bacillus subtilis*	*Escherichia coli*	*Klebsiella Pneumonia*	*Pseudomonas aeruginosa*	*Staphylococcus Aureus*
NiAB	11	16	18	16	15
NiAC	19	15	0	14	18
NiAD	17	14	16	0	16
Control	29	23	30	21	30

ISSN:0971-1260
Vol-22-Issue-4-October-December-2019

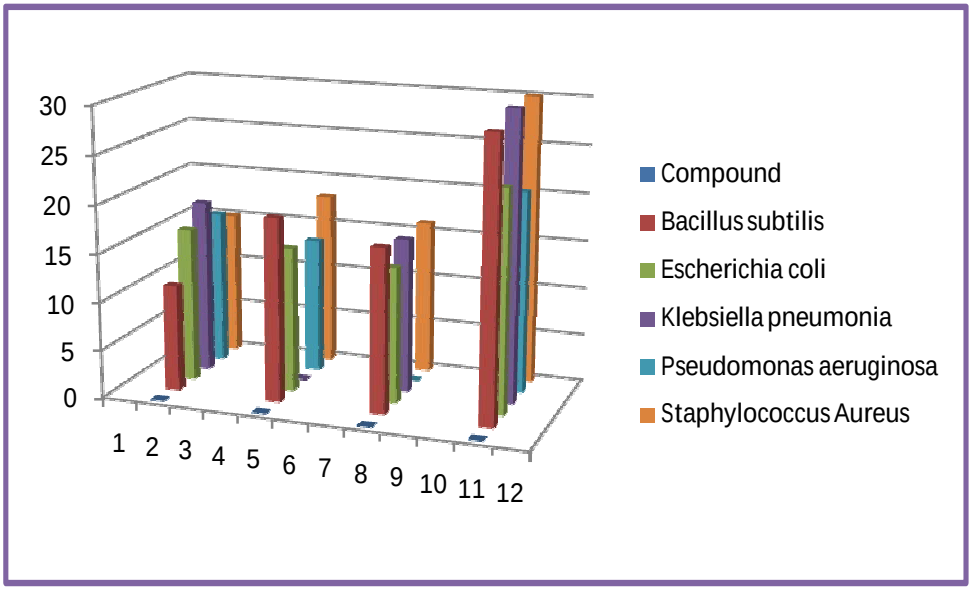


Fig.8. Antimicrobial screening of Nickel complexes by agar well diffusion method

Conclusion

Three Schiff base ligands from essential aminoacids and their nickel complexes have been synthesized and their structure, morphology, physical and chemical properties are determined by IR, UV-Visible, ^{13}C and mass spectral studies. The Schiff base ligands co-ordinate with the metal ions through two azomethine nitrogen, two carboxylato oxygen atoms and two water molecules. Spectral studies suggest octahedral geometry. The DNA cleaveage activity performed by agarose gel electrophoresis using supercoiled pUC19 DNA reveals thatNiAC and NiAD complexes cleave the DNA efficiently from supercoiled form to linear form. The invitro antibacterial activity of the synthesized complexes shows potent antibacterial activity against *B.subtilis* and *S.aureus* species.

References

1. R B Xu, N Zhang, H Y Zhou, S P Yang, Y Y Li, D H Shi, W X Ma, X Y Xu,*Journal of Chemical Crystallography*, **2012**, 42, 928-932.

2. L G Wade, “Organic Chemistry”, 6th Edition, Pearson Prentice Hall, Upper Saddle river, 2005.

ISSN:0971-1260
Vol-22-Issue-4-October-December-2019

3. R J Mandanis, J S Wood, AChandrasekaran, M D Rausch, J C W Chien, *Journal of Organometallic Chemistry*, **2002**, 645, 158-167.

4. I Mhaidat, J A Mergos, SKamilakis, C Kollia, Z Loizos, A Tsolomitis, C T Dervos,*Materials Letters*, **2009**,63,2587-2590.

5. Y G Li, L Pan, Z J Zheng, Y S Li,*Journal of Molecular CatalysisA: Chemical*, **2008**,28757-64.

6. K H Zimmermann, An introduction to Protein Informatics, Kluwer Academic Publishers, London, 2003.

7. M A Neelakantan, R Rusalraj, J Dharmaraja, S Johnsonraja, T Jeyakumar, M S Pillai, *SpectrochimActa A*,**2008**,71, 1599-1609.

8. Z H Chohan, M Arif, M Sarfraz, *Appl.Organomet.chem*,**2007**, 21, 294-302.

9. B K Singh, H Rajour, A Prakash, *SpectrochimActa A*,**2012**,94, 143-151.

10. D D Perrin, W L F Armarego, D R Perrin, Purification of Laboratory Chemicals, Pergamon Press, New York,1980.

11. N Raman, R.Jeyamurugan, A.Sakthivel, L.Mitu, *Spectrochim.Acta A.*, **2010**, 75, 88-97.

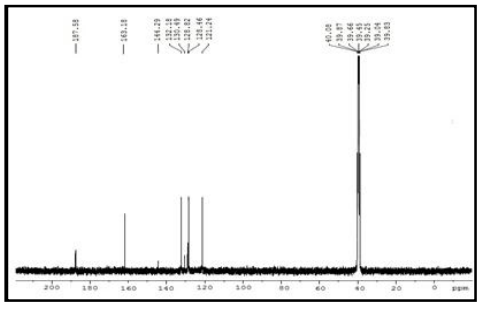
12. W.J.Geary, Coord.chem.Rev., 7(**1971**) 81.

13. N.Raman, S.Sobha, L.Mitu, *Journal of Saudi Chemical Society*,17 (2013) 151-159.

14. Z.H Chohan, M Arif, M A Akhtar, C T Supuran, *Bio inorg.chem.Appl.*, **2006** (2006) 1-13.

15. Z. H. Chohan, A. Scozzafava, C. T. Supuran, *J. Enzyme Inhib.Med.Chem.*, **18** (2003) 259-263.

Supplementary Figures



ISSN:0971-1260
Vol-22-Issue-4-October-December-2019

Fig.S1. ^{13}C NMR spectrum of the Schiff base ligand AB recorded in $CDCl_3$

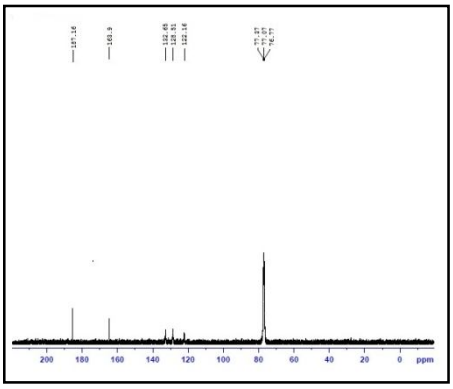


Fig.S2. ^{13}C NMR spectrum of the Schiff base ligand AC recorded in $CDCl_3$

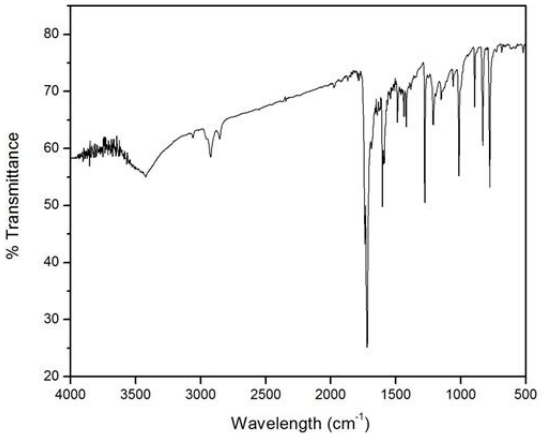


Fig.S3. Vibrational Spectrum of Schiff base ligand (AB)

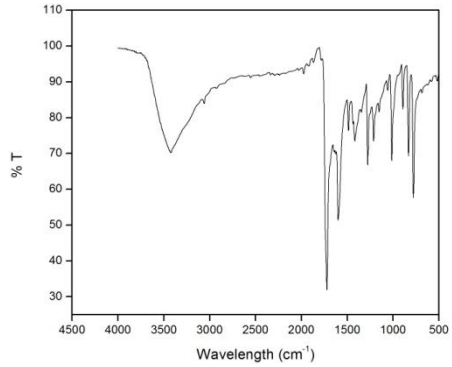


Fig.S4. Vibrational Spectrum of Schiff base ligand (AC)

ISSN:0971-1260
Vol-22-Issue-4-October-December-2019

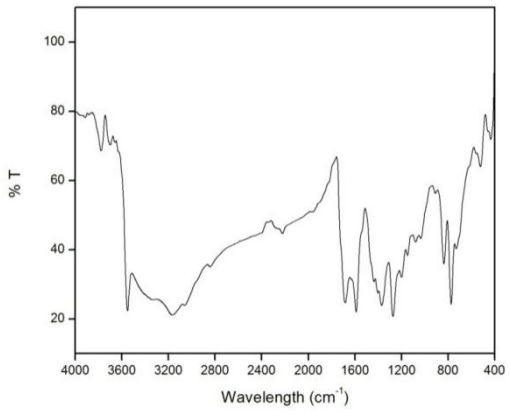


Fig.S5. Vibrational Spectrum of NiAB complex

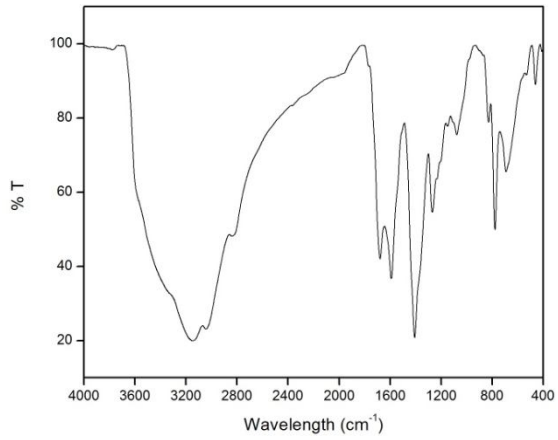


Fig.S6. Vibrational Spectrum of NiAC complex

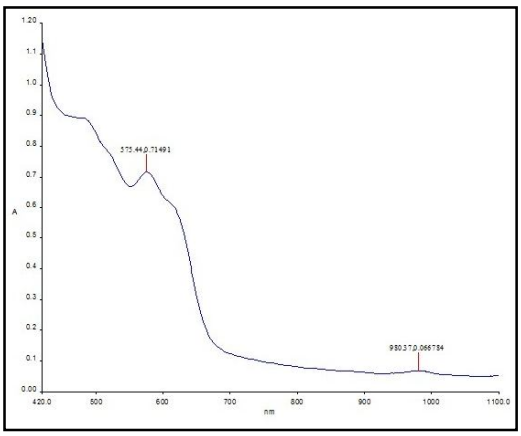


Fig.S7a. Electronic spectrum of NiAB complex(200-1100nm)

ISSN:0971-1260
Vol-22-Issue-4-October-December-2019

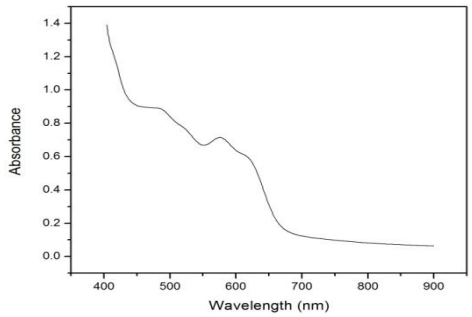


Fig.S7b. Electronic spectrum of NiAB complex (400-900nm)

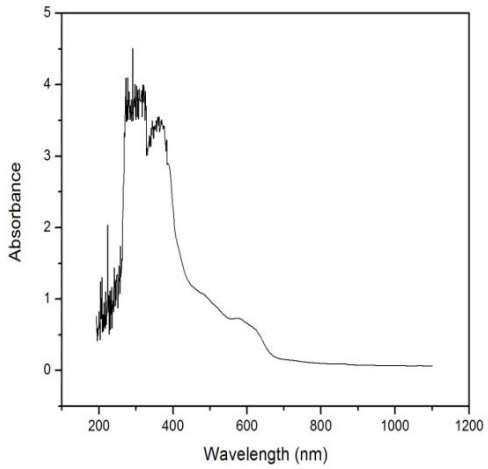


Fig.S8a. Electronic spectrum of NiAD complex(200-1000nm)

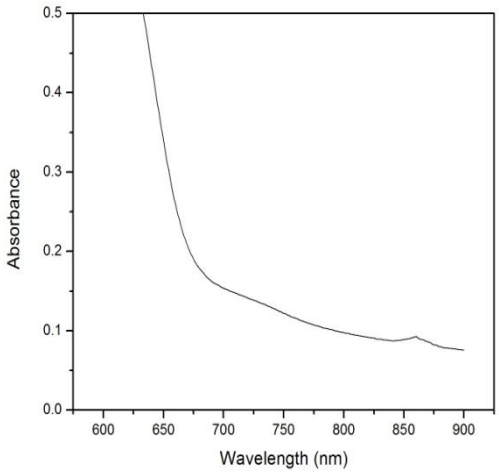


Fig.S8b. Electronic spectrum of NiAD complex (500-800nm)

ISSN:0971-1260
Vol-22-Issue-4-October-December-2019

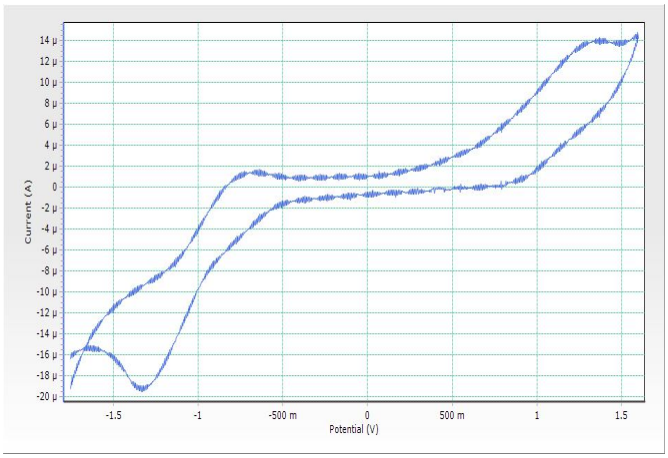


Fig.S9. Cyclic Voltammogram of NiAB complex

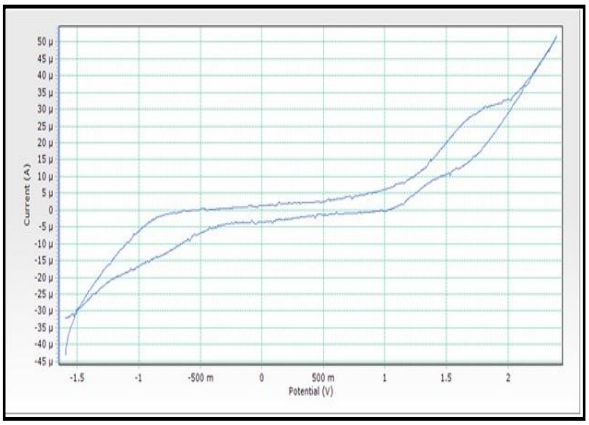


Fig.S10. Cyclic Voltammogram of NiAD complex