Research Journal of Chemical Sciences ISSN 2231-606X
Vol. **3(2),** 35-43, February **(2013)** *Res. J. Chem. Sci.*

Fe(III) Complexes with Schiff base Ligands : Synthesis, Characterization, Antimicrobial Studies

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Available online at: www.isca.in
Received 24th November 2012, revised 1st December 2012, accepted 15th December 2012

Abstract

The synthesis of Fe(III) complexes derived from Schiff base ligands. Obtained by the condensation of o-phenylenediamine, salicylaldehyde and isatin / 2-hydroxy Naphthaldehyde / acetyl acetone is presented. The complexes were characterized by elemental analyses, molar conductance, magnetic susceptibility, IR, Uv-Vis spectral data and thermal analyses. The elemental analysis of the complexes confine to the stoichiometry of the type [ML $(H_2O)_2$] $(OAc)_2$. The complexes were found to be electrolytic in nature on the basis of value of molar conductance. From the spectral datas an octahedral geometry has been proposed for all the complexes. The possible geometries of metal complex were evaluated using 3D molecular modelling picture. The metal complexes have been screened for their antibacterial and antifungal activity.

Keywords: o-phenylenediamine, Salicylaldehyde, 2-hydroxy Naphthaldehyde, Isatin.

Introduction

Schiff bases and their coordination compounds have gained importance recently because of their application as models in biological, biochemical, analytical, antimicrobial system, anticancer, antibacterial and antifungal activities. Studies of new kinds of chemotherapeutic Schiff bases are now attracted the attention of biochemists[1]. Schiff bases of o-phenylenediamine and its complexes have a variety of applications including biological, clinical and analytical[2].

In continuaton of our earlier work on Schiff base complexes[3] in this paper we report the synthesis, characterization and antimicrobial studies of new type tetradentate Schiff base ligands derived from ortho phenylene diamine, salicylaldehyde and isatin / acetyl acetone / 2-hydroxy naphthaldehyde. The ligand has both oxygen and nitrogen donor sites. It coordinates with the metal ion in a tetradentate manner.

Material and Methods

Elemental analyses were performed by using Elementar Vario EL III at STIC, CUSAT, Cochin. The IR spectra were recorded in KBr pellets using Shimadzu FTIR spectrometer (4000 – 400 cm^{-1}). The UV-Vis electronic spectra (200 – 800 nm) were recorded using Lab India 3000^+ double beam spectrophotometer. The magnetic susceptibility of the complexes was recorded at room temperature using Gouy balance. The thermal analysis were performed with a Perkin Elmer (TGS-2 model) thermal analyzer at a heating rate of 10°C / min in the temperature range 40 - 800°C. The AFM images were recorded using Nova software by Multimode Scanning probe microscope (NTMDT, NTEGRA prima, Russia) with cantilever length, width and thickness 135, 30 and 2µm respectively and 0.35 – 6.06N/m force constant. The geometries of metal complex were evaluated using the molecular calculations with Argus lab 4.0.1 version.

Synthesis of Schiff Base Ligand L_1/ L_2/ L_3: A solution of o-phenylenediamine (0.1 mol) in alcohol was added to a mixture of isatin/Acetyl acetone/ 2-hydroxy naphthaldehyde (0.1 mol) and salicylaldehyde (0.1 mol) in 20 ml alcohol. The mixture was refluxed for about 30 minutes. The mixture was cooled in ice. The resulting precipitate was then filtered, washed with ethanol and dried.

Synthesis of metal complexes (ML_1/ M_1L_2/ M_1L_3): To an ethanolic solution of the Schiff Base Ligand L_1/ L2/ L_3 an ethanolic solution of the metal acetate [Ferric Acetate] was added in a molar ratio (1:1). The mixture was refluxed for about 30 minutes. The mixture was cooled in ice. The resulting precipitate was collected by filtration, washed with ethanol and dried.

Antimicrobial studies: The in vitro biological screening effects of the investigated compounds were tested against the bacteria Staphylococcus Aureus, Escherichia Coli and Fungi Candida Albicans. Stock solutions were prepared by dissolving the compounds in DMSO and serial dilutions of the compounds were prepared in sterile distilled water to determine the minimum inhibition concentration (MIC). The nutrient agar medium was poured into Petri plates. A suspension of the tested microorganism (0.5 ml) was spread over the solid nutrient agar plates with the help of a spreader. Different dilutions of the stock solutions were applied on the 10 mm diameter sterile disc. After evaporating the solvent, the discs were placed on the inoculated plates. The Petri plates were placed at low temperature for two hours to allow the diffusion of the chemical and then incubated at a suitable optimum temperature for 30 – 36 hrs. The diameter of the inhibition zones was measured in millimeters[4].

Synthesis of Nano Metal oxides: The transition metal complexes were placed in a silica crucible and ignited in a muffle furnace at 800ºC. The dehydrated mixture undergoes a vigorous, exothermic oxidation reduction reaction. The heat created causes a flame for several minutes, resulting in voluminous and foamy powder product occupying the entire reaction container. The exothermic combustion reaction releases a large amount of heat, which can quickly heat up the system to reach a temperature higher than 1600ºC. The combustion method results in uniform and pure powders of high surface to volume ratio. The size of the metal oxide was determined using Atomic force Microscope.

Results and Discussion

The synthetic routes of the ligands and complexes are presented in scheme 1.

Schiff bases: i. Isatin + o-phenylenediamine + salicylaldehyde $\longrightarrow$ L_1, ii. Acetyl acetone + o-phenylenediamine + sallicylaldehyde $\longrightarrow$ L_2, iii. 2-hydroxynaphthaldehyde+ o-phenylenediamine + sallicylaldehyde $\longrightarrow$ L_{3S}

Metal complexes: $Fe(OAc)_3$+ $L_1/L_2/L_3$ $\longrightarrow$ $Fe(L_1/L_2/L_3)_2$, [Ia, IIa, IIIa], Ia - Isatin Fe complex [$Fe(L_1)$], IIa - Acetyl acetone Fe complex [$Fe(L_2)$], IIIa - 2-hydroxy naphthadehyde Fe complex [$Fe(L_3)$]

Elemental Analysis and Molar Conductance: The metal complexes are insoluble in water and soluble in DMSO, DMF, $CHCl_3$ and acetone and slightly soluble in methanol and ethanol. The analytical data and physical properties of the ligands and complexes are presented in table-1. The data are consistent with the calculated results from the empirical formula of each compound. The analytical data of the complexes confirm the 1:1 metal to ligand stoichiometry.

The molar conductance values measured in DMF solution fall in the range 210 ohm^{-1} cm^{-1} mol^{-1} for Fe complex Ia [$Fe(L_1)$], 200 ohm^{-1} cm^{-1} mol^{-1} for Fe complex IIa [$Fe(L_2)$], 110 ohm^{-1} cm^{-1} mol^{-1} for Fe complex IIIa [$Fe(L_3)$]. The above molar conductance values confirm that the Fe complexes are electrolytes. From the molar conductance values it is evident that the iron complexes Ia [$Fe(L_1)_2$] and IIa [$Fe(L_2)_2$] are 1:2 electrolytes and 1:1 electrolytes for iron complex IIIa[5].

IR spectra: The significant IR bands for the ligands as well as its Fe complex and their tentative assignments are complied and presented in table-2 and figures-1 and 2. In the IR spectrum of the Schiff bases ligands L_1, L_2, L_3 a sharp band observed at 1616 cm^{-1} is assigned to the ν(C=N) mode of the azomethine group. This shifts to lower wave numbers, 1606-1609 cm^{-1} in all the complexes suggesting the co-ordination of the azomethine nitrogen to the metal centres[6]. This is further substantiated by the presence of a new band around 420-463 cm^{-1} assignable to ν(M-N).

The characteristic phenolic ν(O-H) mode due to presence of a hydroxyl group at ortho position in the ligand was observed around 3200-3500 cm^{-1}. A band at ~ 1279 cm^{-1} due to ν(C-O) phenolic group was also observed in the ligand. The disappearance of phenolic ν(OH) band in all the complexes suggests the co -ordination by the phenolic oxygen after deprotonation to the metal ion . This is further supported by the shifting of ν(C-O) phenolic band to lowers wave numbers ~1250 cm^{-1} in the metal complex. The appearance of a new non-ligand band around 500-543 cm^{-1} in all the complexes due to ν(M-O) substantiates it[7].

A strong sharp band observed at 1700 cm^{-1} is assigned to ν(C=O) of isatin and acetyl acetone in ligands L1 and L2. the intensity of this band has not only reduced but has shifted to lower wave numbers in the corresponding metal complexes confirming the involement of the carbonyl group in complexation with metal ion[8].

The presence of co-ordinated water in the Fe complexes is confirmed by the presence of bands around 890 – 928 cm^{-1} [9].

Electronic spectra and magnetic measurements: The electronic spectral data and magnetic moments of the complexes are presented in table- 3 and figures- 3 and 4. The electronic absorption spectra of metal complexes were recorded in DMF in the range 200 – 800 nm.

The electronic spectrum of free Schiff base showed three bands around 240, 350 and 450 nm characteristic of π-π^* and n-π^* transitions[10]. In the metal complexes, this band is shifted to a longer wave length with increasing intensity. This shift may be attributed to the donation of lone pair of electrons of nitrogen of Schiff base to metal ion.

The Fe (III) complexes exhibits bands around 234-253 nm, 324-365 nm and 477-498 nm. The broad intense and poorly resolved bands around 324-365 nm may be assigned to LMCT or MLCT. The high intensity band around 250 nm is of ligand origin assignable to intraligand n-π^*/π-π^* transition. The band around 477-498 nm is assigned to $^6A_{1g} \rightarrow {}^4T_{2g}(G)$[11] transition suggesting octahedral geometry which is confirmed by the magnetic moment value of 5.9 – 5.63 BM[12].

Thermogravimetric Analysis: Thermogravimetric analysis (TGA and DTG) of metal complexes are used to i. get information about the thermal stability of new complexes, ii. decide whether the water molecules are inside or outside the inner co-ordination sphere of the central metal ion and iii. suggest a general scheme for thermal decomposition of chelates.

In the present investigation, heating rates were suitably controlled at 10°C min^{-1} under nitrogen atmosphere and the weight loss was measured from the ambient temperature upto ~ 1000°C. The TGA data are presented in table- 4.

Table–1
Elemental Analysis, Yield, Molar conductivity and Melting point of ligands and complexes

Complex	Emp.Formula	M.Wt	Elemental Analysis, Found (Calculated) %			Conductance (ohm^{-1} cm^{-1} mol^{-1})	Melting point (°C)
			C	H	N		
L_1	$C_{21}H_{15}N_3O_2$	341	-	-	-	-	277
$[Fe(L_1)]$	$C_{25}H_{24}N_3O_8$. Fe	549.85	53.9 (54.6)	4 (4.4)	7.2 (7.6)	210	> 360
L_2	$C_{18}H_{18}N_2O_2$	294	-	-	-	-	298
$[Fe(L_2)]$	$C_{22}H_{27}N_2O_8$. Fe	502.85	51 (52.5)	5 (5.4)	5.2 (5.6)	200	300
L_3	$C_{24}H_{17}N_2O_2$	351	-	-	-	-	326
$[Fe(L_3)]$	$C_{26}H_{23}N_2O_6$.Fe	514.85	59 (60.6)	4 (4.5)	5 (5.4)	110	> 360

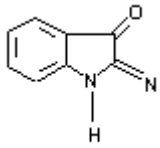
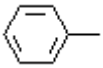
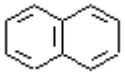
Table-2
IR spectral data for ligands and their metal complexes

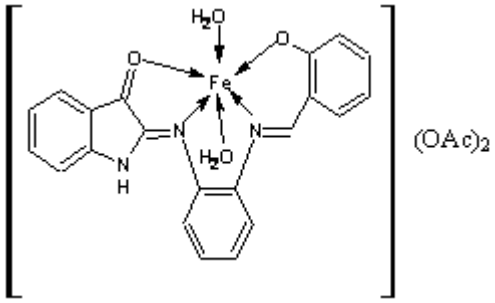
Complex	ν(C=O)	ν(C=N)	ν(OH)	ν(M-N)	ν(M-O)	ν (NH)	ν(C-O)	ν(H_2O)
L_1	1707.06	1616.40	3319.60	-	-	3059.20	1279.81	-
$[Fe(L_1)]$	1700.31	1607.72	-	457.14	540.09	3040.00	1252.81	890.81
L_2	1700.31	1614.47	3228.95	-	-	-	1275.95	-
$[Fe(L_2)]$	1692.59	1606.76	-	463.90	537.19	-	1251.84	916.72
L_3	-	1616.40	3531.76	-	-	-	1279.81	-
$[Fe(L_3)]$	-	1619.29	-	463.90	555.50	-	1255.70	911.40

Table-3
Electronic Spectral and Magnetic moment data for the ligands and their complexes

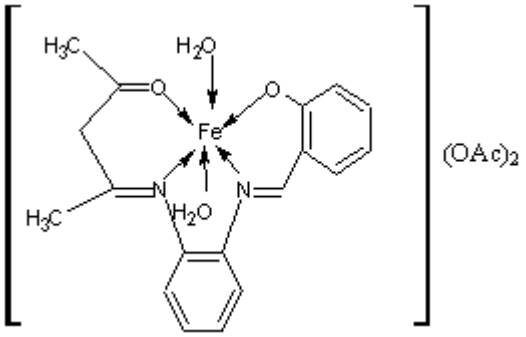
Ligand/ Complex	Absorbance nm	ν/cm^{-1}	Assignment	Geometry	Magnetic moment (BM)
L_1	350	28571	π - π*	-	-
	425	23529	n - π*		
L_2	338	29585	π - π*	-	-
	445	22471	n - π*		
L_3	355	28169	π - π*	-	-
	479	20876	n - π*		
$[Fe(L_1)]$	253	39525	INCT n - π* / π - π*	Octahedral	5.63
	365	27397	MLCT		
	498	20080	$^5T_{2g}$ (F) → ^5Eg		
$[Fe(L_2)]$	234	42735	INCT n - π* / π - π*	Octahedral	5.72
	350	28571	MLCT		
	477	20964	$^5T_{2g}$ (F) → ^5Eg		
$[Fe(L_3)_2]$	324	30864	MLCT	Octahedral	5.9
	492	20325	$^5T_{2g}$ (F) → ^5Eg		

Table-4
Thermal Analysis data for metal complexes

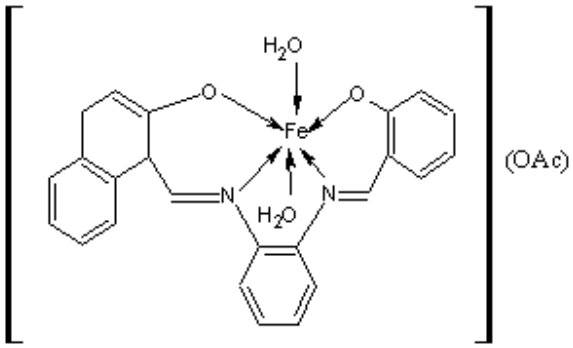
Complex	Decomposition Temp°C	Lost fragment	Weight loss %	
			Experimental	Theoretical
[Fe(L_1)]	160-240	$2H_2O$	6.5	6
	260-400	$2CH_3COO$	22.9	23
	650 – 750		20.66	20
	> 750	Residue FeO	24.23	23
[Fe(L_2)]	156 – 230	$2H_2O$	7.2	6.5
	260-430	$2CH_3COO$	22.27	20
	550-700		16.27	17
	> 700	Residue FeO	38.37	40
[Fe(L_3)]	150-270	$2H_2O$	7.3	8
	313 – 360	CH_3COO	12.3	10
	560-690		30	30
	> 800	Residue FeO	35.17	35



[Fe (L_1) (H_2O)$_2$] (OAC)$_2$



[Fe (L_2) (H_2O)$_2$] (OAC)$_2$



[Fe (L_3) (H_2O)$_2$] (OAC)

Scheme-1
Based on the above results probable structures have been proposed

The TGA curve of the Fe (III) complexes (Ia/IIa/IIIa) figure-5 showed a rapid first step decomposition around 160-250°C with 6.5 – 7.3 % mass loss (calculated 6 – 8%) indicating the loss of two coordinated water molecules. The complex Ia [Fe(L_1)] shows a weight loss of 22.9% at a temperature range 260-400°C which corresponds to the removal of two acetate groups. A weight loss of 20.66% is observed in the temperature range 650 - 750°C which suggests the elimination of isatin moiety. In the Fe complex (IIa) [Fe(L_2)] / (IIIa) [Fe(L_3)] a weight loss of 22.72 and 12.3% at temperature range 260-430°C and 313-360°C corresponds to the removal of two acetate and one acetate groups respectively. Similarly at a temperature of 550-700°C and 560-690°C a phenyl / naphthyl moiety is eliminated corresponding to a weight loss of 16.27 % and 30% respectively. The decomposition is completed at ~ 800°C leading to the formation of the stable metal oxide FeO (Ia [Fe(L_1)]– 24.3%, IIa [Fe(L_2)] – 38.37%, IIIa [Fe(L_3)] – 35.17%).

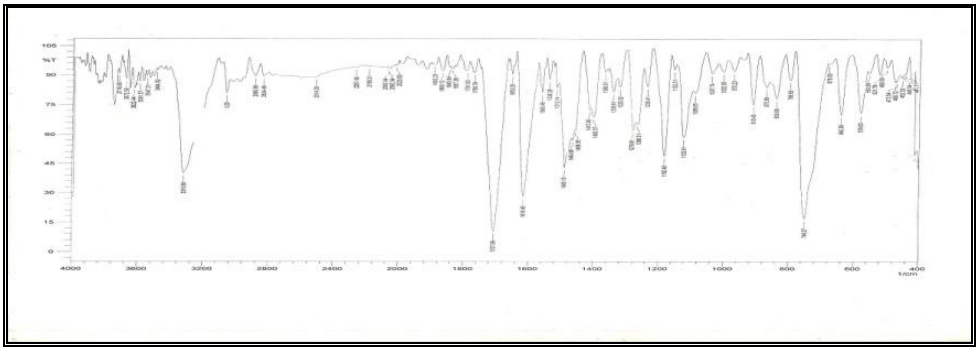


Figure-1
IR Spectrum of Schiff base L_1

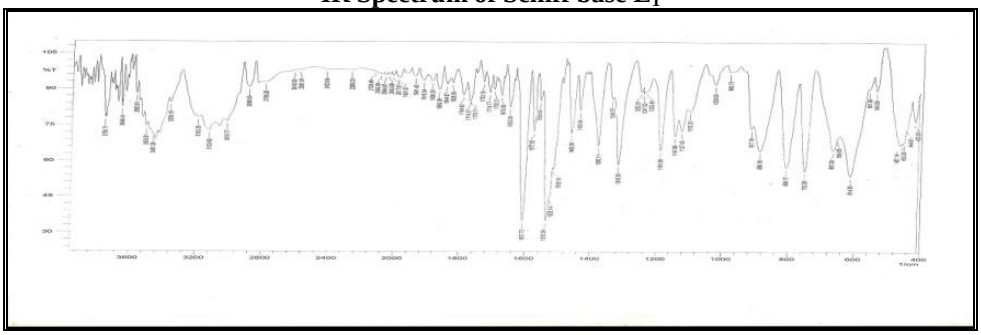


Figure-2
IR Spectrum of complex [Fe.L_1 (H_2O)$_2$] (OAc)$_2$

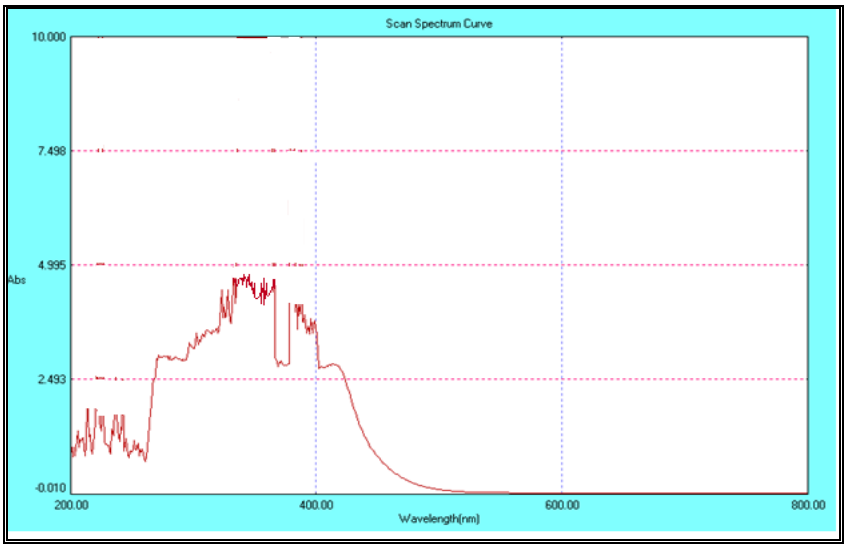


Figure-3
Electronic spectrum of Schiff base L_1 [Fe.L_1 (H_2O)$_2$ OAc)$_2$]

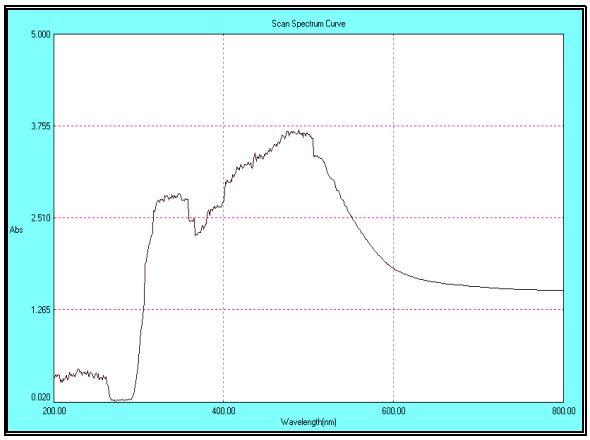


Figure-4
Electronic Spectrum of complex

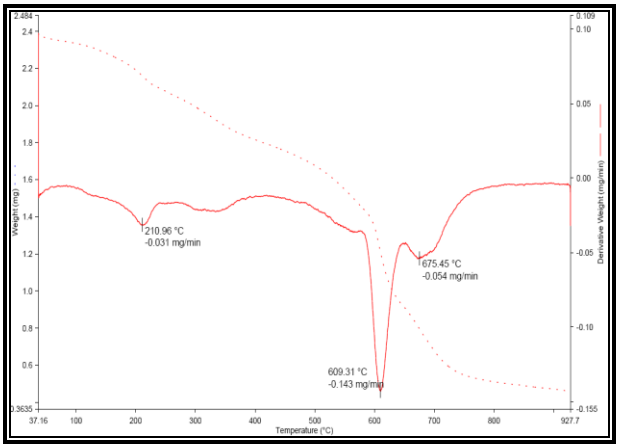


Figure-5
Thermogravimetric analysis of complex [Fe.L_1 (H_2O) $_2$ ($OAc)_2$]

Molecular Modelling: Molecular modeling of the studied complexes reveals minimum energy values associated with the octahedral geometry. This is in a good agreement with the experimental results and confirmed the expected structure. The possible geometries of metal complexes were evaluated using the molecular calculations with Argus lab 4.0.1 version software. The metal complexes were built and geometry optimization was done using this software. The molecular modelling pictures and the energies of the metal complexes are shown in figures-6 and 7.

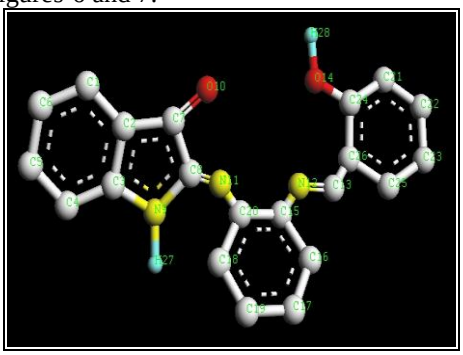


Figure-6
Schiff base L_1 Energy 93.61 kcal/mol

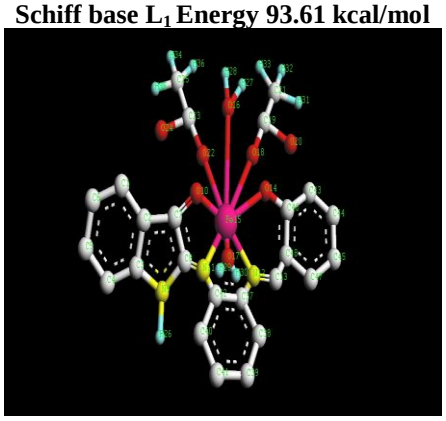


Figure-7
Metal complex [Fe. $L_1(H_2O)_2$] $(OAc)_2$ Energy 651.52 kcal/mol

The details of important bond lengths as per 3D structure of Fe(III) complex (Ia) [figure 10] are given in table-5. These values are obtained as a result of energy minimization of Fe (III) complex in Argus lab 4.0.1 version software.

The obtained bond lengths of the ligand L_1 based on the software are between C(7)-O(10) and O(14)-C(46) 1.3654ºA, C(8)-N(11) and N(12)-C(13) 1.4567ºA and N(11)-C(42) and N(12)-C(13) 1.5348ºA. Based on the values from the table 5, it is observed that when these ligand are coordinated with Fe metal ion there is an increase in the bond length between the mentioned atoms, which confirms the coordination of azomethine group through nitrogen [N(11) and N(12)] and through phenolic oxygen [O(10) and O(14)]. When the atoms are coordinated with the metal ion by donating a lone pair of electron there is a decrease of electron density on the coordinating atoms, hence bond length increases in metal complexes. This supports the proposed octahedral geometry around the iron metal ion[13].

Table-5
Data from Molecular modelling of [Fe(L_1)] complex

S. No.	Bonded atoms	Bond length
1	3(C) - 9(N)	1.356681
2	7(C) - 10(O)	1.36541
3	11(N) - 42(C)	1.434808
4	14(O) - 46(C)	1.36541
5	9(N) - 26(H)	1.062577
6	10(O) - 15(Fe)	2.077424
7	11(N) - 15(Fe)	2.079850
8	12(N) - 15(Fe)	2.079850
9	14(O) - 15(Fe)	2.077424
10	15(Fe) - 16(O)	2.077424
11	16(O) - 27(H)	1.033746
12	15(Fe) - 17(O)	2.077424

Anti Bacterial studies: The antimicrobial activity of the Fe complexes was studied against two pathogenic bacterial strains

(ie) one gram positive (staphylococcus Aureus) and one gram negative (Escherichia Coli) bacteria and one fungal strain (Candida Albicans) figures-8 - 10. Antibacterial and antifungal potential of Fe complexes were assessed in terms of zone of inhibition of bacterial and fungal growth. The results of the antifungal and antibacterial activities are presented in table-6. The minimum inhibitory concentrations (MIC) were calculated as the highest dilution showing complete inhibition of the tested strains and are reported in table-7.

Figure-8
Zone of inhibition – bacteria *S.Aaureus* image of [Fe.L_1 $(H_2O)_2$] $(OAc)_2$

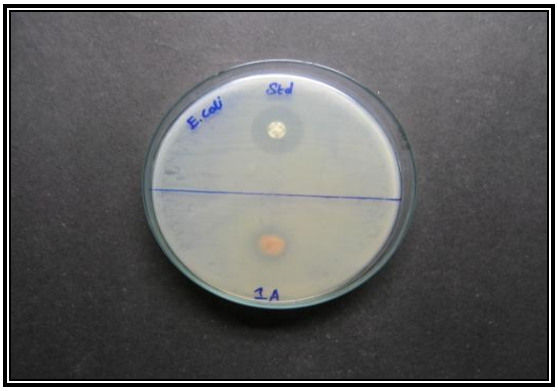


Figure-9
Zone of inhibition – bacteria *E.Coli* image of [Fe.L_1 $(H_2O)_2$] $(OAc)_2$

Figure-10
Zone of inhibition – fungi *C.Albicans* image of [Fe.L_1 $(H_2O)_2$] $(OAc)_2$

The complexes were effective against both bacteria and fungi. The iron complex was better against both the gram positive and gram negative bacteria and fungi when compared with the standard ciprofloxacin and Clotrimazole. Growth of bacterial pathogens on each concentration was checked to determine the minimum concentration that inhibits the growth of the organism. It is evident from the table that the MIC value for iron complex was 125 µg/ml against *S.Aureus* and 62.5 µg/ml against *E.Coli*. Likewise the MIC value for fungi pathogen shows 62.5 µg/ml for iron complexes (table-8).

Table-6
Antibacterial and Antifungal activity data of Schiff base metal complexes

Microorganism		**$[Fe(L_1)_2(H_2O)_2]OAc$**	**STD(50 µg/disc) (mm)**
Bacteria (Ciprofloxacin)	*E.Coli*	15	20
	S.Aureus	12	25
Fungi (Clotrimazole)	*C.albicans*	20	18

Table-7
Determination of MIC for antibacterial and antifungal activity

Microorganism		500 µg/ml	250 µg/ml	125 µg/ml	62.5 µg/ml	31.25 µg/ml	15.62 µg/ml	7.8 µg/ml
Bacteria	S.Aureus	-	-	-	+	+	+	+
	E.coli	-	-	-	-	+	+	+
Fungi	*C.Albicans*	-	-	-	-	+	+	+

Table-8
Antibacterial and Antifungal activity: MIC values

Microorganism		MIC value
Bacteria	S.Aureus	125 µg/ml
	E.coli	62.5 µg/ml
Fungi	*C.Albicans*	62.5 µg/ml

Determination of size of nano metal oxide: The size of the metal oxide was determined by Atomic force Microscopy. AFM provides a 3D profile of the surface on a nanoscale by measuring forces between a sharp probe (<10 nm) and the surface at very short distance (0.2 – 10 nm probe – sample separation. The 2D AFM images of the metal oxide of Ia are shown in figure-11 and the 3D images in figure-12. From the figures it is evident that the size of the synthesized metal oxide is in the range (~ 0 - 20 nm). From the 2D AFM images of the metal oxide of IIa and IIIa the size of the metal oxides was found to be in range 0-100 nm.

Conclusion

In this paper we have reported the synthesis of Schiff base ligands derived from acetyl acetone / isatin / 2-hydroxy naphthaldehyde with o-phenylenediamine and salicylaldehyde and their Fe(III) complexes have been synthesized using the Schiff base ligands.

The ligands and complexes were characterized by spectral and analytical data. Based on these data an octahedral geometry has been assigned to the Fe(III) complexes. Molecular modelling has been performed for the complexes using Argus 4.0.1 software. The metal complexes were converted to their corresponding nano metal oxides, the 2D and 3D AFM pictures of the oxides confirm their size to be in the range 0 - 20 nm. The antimicrobial studies carried out with the complexes confirm that they are good anti bacterial and antifungal agents with their MIC values being 125 and 62.5 µg / litre.

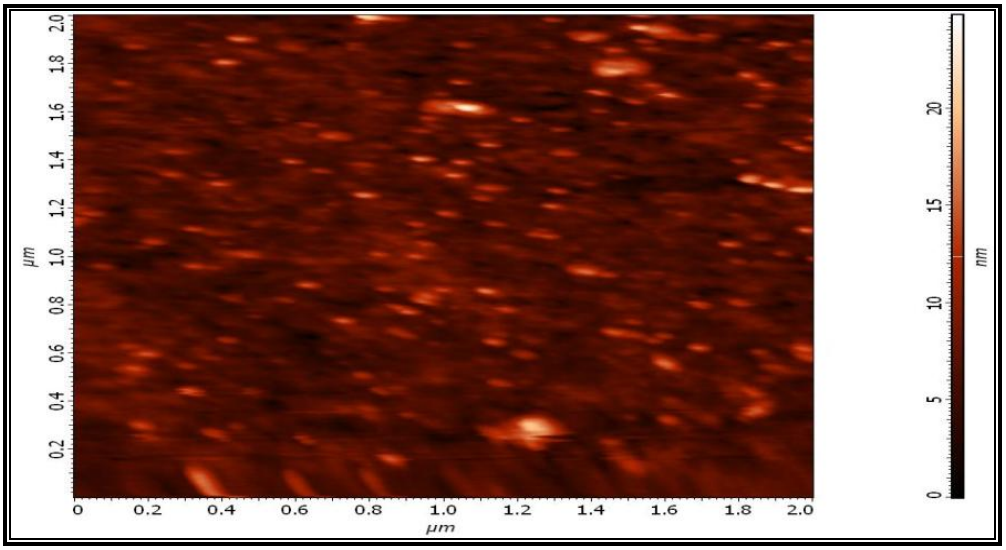


Figure-11
AFM topographic images of Fe oxide of $[Fe(L_1)_2(H_2O)_2](OAc)_2$ showing 2D configuration in the material. The scanned area is 0.2 µm x 0.2 µm.Size of the particle = 10nm-20-nm

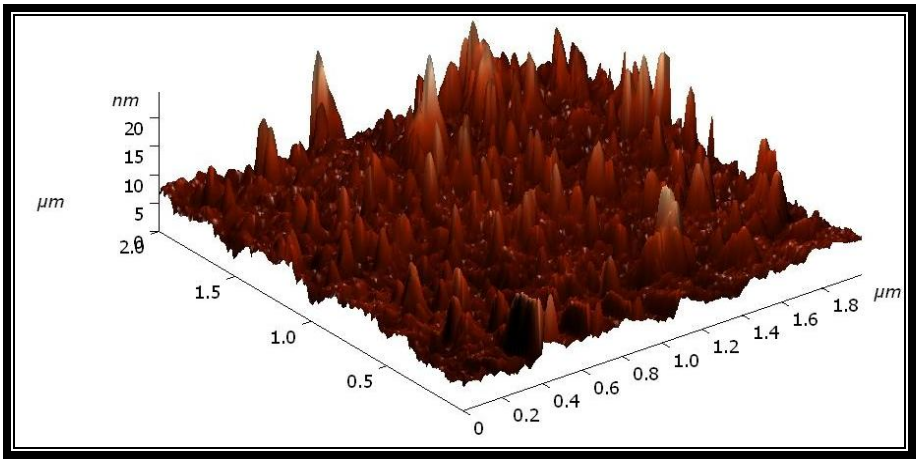


Figure-12
AFM topographic images of Fe oxide of $[Fe(L_1)_2(H_2O)_2](OAc)_2$ showing 3D configuration in the material, The scanned area is 0.2 µm x 0.2 µm. Size of the particle = 10nm-20-nm

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