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### Synthesis and Characterization of Mixed Ligand Complexes of Fe (III) with 2-hydroxyacetophenone and Substituted Salicylaldehydes or β-diketones

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#### ABSTRACT

Some new mixed ligand complexes of Fe (III) of the type [Fe(L)<sub>2</sub>L'], (where HL=2hydroxyacetophenone and HL'=Salicylaldehye,5-bromosalicylaldehyde,5chlorosalicylaldehyde, 5-nitrosalicylaldehyde,2-hydroxy-1-naphthaldehyde, pentane-2,4-1-phenylbutane-1,3-dione or 1,3-diphenylpropane-1,3-dione) have dione, been synthesized by the condensation reaction of iron(III) chloride with a mixture of two different ligands in 1:2:1 molar ratios. The mode of bonding and geometry of the compounds were determined through physicochemical and spectroscopic methods. Biological activity of ligands and metal complexes was performed against gram positive bacterial strain Staphylococcus aureus and gram negative bacteria Escherichia coli. Keywords: Mixed ligand complexes, 2-hydroxyacetophenone, [Fe(2-hap)<sub>2</sub>(dbzm)], conductance of mixed ligands, antibacterial activity.

#### INTRODUCTION

In recent decades, chemistry of mixed ligand complexes has played a central role in the conceptual development of modern chemistry. Mixed ligand complexes are useful in the storage and transport of active substances through membrane Chandra and Tyagi (2008). These are helpful in activation of enzymeand analysis of semiconductor material Panda and Mohaptra (1984). Various oxidation states of iron and the wide applications of its mixed ligand complexes in biological and industrial fields are gaining popularity in the chemistry now a day's Chen et al. (2010).

A lot of work has been done to synthesize mixed ligand complexes of transition metals. Rani et al.(1994) reported Mn(II), Fe(III), Co(II), Ni(II), Cu(II) and Zn(II) complexes of a new multidentate oxygen nitrogen donor, bis(N-salicylidene)-2,3-dihydrazino-1,4-quino xaline. Few mixed ligand complexes of Fe(III) with  $\alpha$  -diketones and nitrogen or phosphorus

ligands been reportedGnanasoundari Natarajan (2005). containing have and Revanasiddappa et al. (2009) have been synthesized Fe(III) complexes with lansoprazole 2-{3-methyl-4-(2,2,2-fluroethane)-2-pyridyl)methyl}sulphonylbenzdrug, that is imidazole.sharma et al. (2005) have been synthesized Cr(III), Mn(III), Fe(III) and Co(III) complexes of hydrazones (derived from substituted benzoyl hydrazides) and benzyl  $\alpha$ monoxime. Amani et al. (2007) reported Mixed ligand complexes of the type [Fe(bipy)Cl<sub>3</sub> (DMSO)] and [Fe(phen)Cl<sub>3</sub> (DMSO)] (Where, bipy=2,2'-bipyridine, phen=1,10-phenanthroline and DMSO=dimethyl sulfoxide) by the treatment of [Fe(bipy)Cl<sub>4</sub>](bipy.H) and [Fe(phen)Cl<sub>4</sub> ][phen.H] with DMSO in methanolic solution respectively. Foca et al. (2004) have synthesized and characterized some Cr (III), Fe (III) and Zr (IV) complexes with substituted (2-hydroxy-4methoxyphenyl) phenylmethanone.

In this paper we report some new mixed ligand complexes of Fe(III) because of the superiority in biocompatibility of iron metal over other transition metalsNanami et al. (2005) . Resulting complexes are characterized by different techniques as -UV, IR, FAB mass, molar conductivity and antibacterial activity. On the basis of these studies coordination behavior and geometry of new mixed ligand complexes were discussed.

#### MATERIALS AND METHODS

#### Materials

5-nitrosalicylaldehyde (Merck), 5-Bromosalicylaldehyde (Aldrich), 5-chlorosalicylaldehyde (Aldrich), 1-phenylbutane-1,3-dione (sisco-chem), 2-hydroxy-1-naphthaldehyde (Merck) and 1,3-diphenylpropane-1,3-dione (Sisco-chem) were purified by recrystallization from butanol prior to use. Salicylaldehyde (Merck), 2-hydroxyacetophenone (Merck), Pentane-2,4-dione (JohnBaker) and butanol were purified by distillation. Iron (III) chloride A.R. was used as supplied.

#### Analytical methods and physical measurements

Iron was estimated volumetrically by EDTA using Eriochrome black-T as an indicatorvogel (1996). Carbon and hydrogen analyses were carried out on a HeraeusCarloErba 1108 instrument. Molar conductances were measured at room temperature in DMSO by a Chino make digital conductivity meter (model no-NDC-736). Magnetic measurements were carried out using MSB MK1 Magnetic susceptibility balance, Sherwood Scientific. Electronic spectra were recorded in DMSO in the range of 200nm-800nm on a SYSTRONICS UV-VIS Spectrophotometer. Infrared spectra of the complexes were recorded in the region 4000-400cm<sup>-1</sup> on a SHIMADZU-JAPAN8400S FTIR spectrophotometer using KBr pellets. The FAB mass spectra were recorded on a Jeol SX102/DA-600 mass spectrometer/Data System using Argon/Xenon (6kV, 10mA) as the FAB gas. The accelerating voltage was 10kV and spectra were recorded at room temperature. m-Nitrobenzyl Alcohol(NBA) was used as the matrix.

#### Antibacterial screening

The in vitro antibacterial activities of the ligands and metal complexes were tested by using Muller Hinton agar by well diffusion method against a gram negative bacterial strain *Escherichia coli*(MTCC 443) and a gram positive bacterial strain *Staphylococcus aureus* (MTCC 96). The bacterial strains grown on nutrient agar at 37°C for 18 hours were suspended in a saline solution (0.85% NaCl) and adjusted to a turbidity of 0.5 McFarland standards [10<sup>8</sup> colony forming units (CFU)/ml].

The suspension was used to inoculate 90 mm diameter Petri plates. Wells (6 mm diameter) were punched in the agar with the help of a sterile metallic borer and filled with 100 µl of the test extract of the concentration 10 mg/mL. The dissolution of the organic extracts (Methanol) was added by DMSO which did not affect the growth of microorganisms, in accordance with our control experiments. Plates were incubated in air at 37°C for 24 hours. Antibacterial activities were evaluated by measuring diameters of the inhibition zone in millimeters carefully. DMSO was taken as control for the methanol extracts and ciprofloxacin (10mg) was used as a reference standard antibiotic drug.

#### Synthesis of mixed ligand complexes of Fe(III)

(a) Synthesis of Mixed Ligand Complexes of Fe (III) with 2-hydroxyacetophenone and  $\beta$  -diketones.

(b) Synthesis of Mixed Ligand Complexes of Fe (III) with 2-hydroxyacetophenone and substitutedsalicylaldehydes.

## (a) Synthesis of mixed ligand complexes of Fe(III) with 2-hydroxyacetophenone and $\beta$ -diketones

To a butanolic solution of FeCl<sub>3</sub> (5.24 mmol, 0.851g in 10 ml *n*-butanol), butanolic solution of 2-hydroxyacetophenone (1.34ml) and 1,3-diphenylpropane-1,3-dione (1.10g in 10 ml *n*-butanol) were added with constant stirring. The reaction mixture was stirred for about 30-40 minutes. No ppt was obtained. Then, added 5% aqueous sodium hydroxide solution (~4 ml) drop wise to the above reaction mixture to raise the pH upto ~6.0. Stirred the solution for 3-4 hrs and then reflux the reaction mixture on heating mental for 4-5 hours, the ppt began appear. After refluxing, the reaction mixture was kept on room temperature. The ppt was settled down. Filter the solution, washed with butanol and dried properly under reduced pressure.

A similar method was adopted to synthesize mixed ligand complexes of Fe(III) with Salicylaldehye, 5-bromosalicylaldehyde, 5-chlorosalicylaldehyde, 5-nitrosalicylaldehyde, 2-hydroxy-1-naphthaldehyde, Pentane-2,4-dione, and 1-phenylbutane-1,3-dione.

#### **RESULTS AND DISCUSSION**

The reactions of iron (III) chloride with 2-hydroxyacetophenone and  $\beta$ -diketones

In 1:2:1 molar ratios result in the formation of mixed ligand complexes of iron, scheme 1. Similarly in scheme 2, mixed ligand complexes of Fe(III) were synthesized with

2-hydroxyacetophenone and Salicylaldehye, 5-bromosalicylaldehyde, 5chlorosalicylaldehyde, 5-nitrosalicylaldehyde.the mixed ligand complexes of Fe(III) with 2hydroxyacetophenone and 2-hydroxy-1-naphthaldehyde represent in scheme 3.

#### Analytical and Physical measurements of synthesized complexes

The resulting mixed ligand complexes are obtained in 32-57% yields as reddish brown or blackbrown solid. The data of C, H and Fe analyses agree well with the calculated values corresponding to the respective complexes. The complexes decomposed at high temperature on heating. All the physical data are shown in Table I.

#### Magnetic moments

The  $\mu_{eff}$  values for the complexes are observed in the range 5.32 to 5.89 B.M. as expected for five unpaired electrons.

According to the  $\mu_{eff}$  value of these complexes the metal present in d<sup>5</sup> electronic configuration. Thus, all the complexes are high spin paramagnetic complexesBhattacharjee et al. (2010). It lies within the octahedral range which is very close to spin value 5.90 B.M. as the ground term is  ${}^{6}A_{1}g$  and thus supports the octahedral geometry.



Scheme 1. Synthesis of mixed ligand complexes of Fe(III) with 2-hap and  $\beta$ -diketones



Scheme 2. Synthesis of mixed ligand complexes of Fe(III) with 2-hap and substituted salicylaldehydes



Scheme 3. Synthesis of mixed ligand complexes of Fe(III) with 2-hap and 2-hnp (2-hnp=2-hydroxy-1-naphthaldehyde)

S.N	Complex	Colour	Decomp.	Analysis % Found		Molar		
0.	Molecular Formula	Yield%	Temp.	(calculated)		Cond.(mh	$\mu_{eff}/B$	
	MolecularWeight		(°C)	Fe	С	Н	0	.M )
							cm²mol⁻¹)	
1.	[Fe(2-hap) <sub>2</sub> (sal)]	Brown	285	12.48	61.76	4.28	8.2	5.78
	C <sub>23</sub> H <sub>19</sub> O <sub>6</sub> Fe	37.67		(12.12)	(61.23)	(4.98)		
	447.24 g/mol							
2.	[Fe(2-hap) <sub>2</sub> (5-Br sal)]	Blood	298	10.61	52.50	3.44	5.76	5.65
	C <sub>23</sub> H <sub>18</sub> O <sub>6</sub> Br Fe	Red		(10.32)	(52.12)	(3.61)		
	526.13 g/mol	39.67						
3.	Fe(2-hap) <sub>2</sub> (5-Cl sal)]	Reddish	289	11.59	57.34	3.76	6.4	5.69
	C <sub>23</sub> H <sub>18</sub> O <sub>6</sub> Fe Cl	black		(11.34)	(57.87)	(3.89)		
	481.73 g/mol	41.78						
4.	Fe(2-hap)₂(5-nitro sal)]	Brown	296	11.33	56.12	3.68	7.45	5.32
	C <sub>23</sub> H <sub>18</sub> O <sub>8</sub> NFe	43.89		(10.65)	(56.67)	(3.78)		
	492.24 g/mol							
5.	[Fe(2-hap)₂(2-hnp)]	Black	316	11.22	65.21	4.25	6.1	5.35
	$C_{27}H_{21}O_{6}Fe$	41.66		(11.35)	(65.89)	(4.67)		
	497.30g/mol							
6.	Fe(2-hap)₂(acac)]	Blackish	312	15.11	68.28	5.72	5.12	5.87
	$C_{21}H_{21}O_6$ Fe	Brown		(15.45)	(68.98)	(5.65)		
	362.39 g/mol	42.87						
7.	Fe(2-hap) <sub>2</sub> (bzac)]	Reddish	293	11.31	64.19	4.32	7.56	5.76
	C <sub>26</sub> H <sub>24</sub> O <sub>6</sub> Fe	black		(11.54)	(64.34)	(4.65)		
	486.23 g/mol	46.17						
8.	Fe(2-hap) <sub>2</sub> (dbzm)]	Red	301	10.16	67.77	4.58	11	5.89
	$C_{31}H_{25}O_{6}Fe$	58.98		(10.34)	(67.23)	(4.24)		
	549.37 g/mol							

Table 1. Analyses and Cl	naracterization of Mi	xed Ligand Con	mplexes [Fe (	2-hap)₂(dbzr	n)]

These are insoluble in water or most of the organic solvents like methanol, benzene and carbon tetrachloride but soluble in DMSO and DMF.

#### Infra-red spectra

The IR spectra of all complexes reexhibited absorption bands with the appropriate shifts due to complex formation. In the IR spectra of the mixed ligand complexes the absorption bands at 1635-1655 cm<sup>-1</sup> may be assigned to v(C=O)and those at 1165-1220 cm<sup>-1</sup> due to v(C-O) respectively.

In free 2-hydroxyacetophenone, 2-hydroxy-1-naphthaldehyde, salicylaldehyde, benzoyl acetone, 5-bromosalicylaldehyde, acetylacetone bands appeared due to v(C=O) at 1650, 1806, 1680, 1650, 1670 and 1640 cm<sup>-1</sup> respectivelyNakamoto and Martell (1966). Thus these bands shift to lower wave number side in the complexes supporting the coordination of these ligands with the metal ions. Bellamy and Beecher (1954) initially attributed the 1580 and 1520 cm<sup>-1</sup> bands to C=O and C=C stretching respectively. The assignments were reversed by Nakamoto et al (1966) on the basis of force constant calculations. Mikami et al. (1967) supported the later view although the two modes were shown tobe coupled slightly with each other. The recorded IR spectrum of [Fe (2-hap)<sub>2</sub>(dbzm)] is reproduced in Figure-1. Strong absorption bands in the region 1550-1510 cm<sup>-1</sup> may be attributed to coordinated to v(C=C) and due to coordination of the Carbonyl oxygen of the ligands to the central metal ion, negative Shift of this band to a value 1656-1620 cm<sup>-1</sup>occurs in the complex. Weak absorption bands in the region 500-450 cm<sup>-1</sup>, which are absent in the free ligands, may be attributed to v (Fe-O) vibrations.



#### **Electronic spectra**

UV-vis spectrum of the  $[Fe(2-hap)_2(dbzm)]$  was recorded in DMSO as solvent(Reference Solvent was taken DMSO) is shown in figure 2. The electronic spectra of Fe(III) complexes exhibit three bands. First intense absorption band in the range of 300-380 nm because of assigned to p-p\* intra-ligand transition. Second relatively less absorption band at 380-450 is due to  $t_{2g}$  - p\* transition (metal to ligand charge transfer) and third band at 450-550 nm because of the p-e<sub>g</sub> transition (ligand to metal charge transfer) respectively. Existence of these three bands confirms the formation of the mixed complex Fe(2-hap)<sub>2</sub>(dbzm)]Feltham and Hayter (1964).

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Figure 2. UV-vis spectrum of the [Fe(2-hap)<sub>2</sub>(dbzm)]

#### FAB mass spectra

FAB mass spectrum of the complex  $[Fe(2-hap)_2(dbzm)]$  has been recorded and fragmentation pattern of complex during the present course of investigation given in figure-3 and m/z values of the peaks along with their intensities with respect to base peak are shown in table II.

## TABLE II. Mass spectral data of mixed ligand complexes of Fe (III) (m/z values and relativeAbundances) [HL= 2-hap, HL' = dbzm]

lon	Fe(2-hap) <sub>2</sub> (dbzm)
FeL <sub>2</sub> L' <sup>+</sup>	551(27.1%)
FeL2' <sup>+</sup>	502(100%)
FeL2 <sup>+</sup>	324.7(44%)
FeL⁺	195 (22.47%)
FeL' <sup>+</sup>	268(74.54%)
Fe <sub>2</sub> LL <sub>2</sub> <sup>+</sup>	698(18.36%)
Fe <sub>2</sub> L <sub>2</sub> L' <sup>+</sup>	592(59.27%)
FeL <sub>3</sub> <sup>+</sup>	469(49.63%)

The Complex shows sharp base peak at m/z(502, 100 %) which may be formed due to the loss of 2-hydroxyacetophenone ligand moiety.

Formation of various species can be explained due to the formation of FeL<sup>+</sup> and FeL<sup>+</sup> species as a result of the loss of one or the other ligand moiety and shown with the help of the following reactions-

$\operatorname{FeL}_{2} L^{+} \xrightarrow{-L} \operatorname{FeL}_{2}^{+} \xrightarrow{-L} \operatorname{FeL}^{+}$	(1)
$\operatorname{FeL}_{2} L^{+} \xrightarrow{-L} \operatorname{FeLL}^{+} \xrightarrow{-L} \operatorname{FeL}^{+} $	(2)

A peak is observed due to molecular ion  $[Fe (2-hap)_2(dbzm)]^+$  at m/z(551, 27.1%). Quite intense peak is seen at m/z(324.7, 44%) due to  $[FeL_2^+]$  which is formed by the loss of dibenzoylmethane ligand moiety.

The peak corresponding to  $\text{FeL}^+$  which would be resulted by the loss of dibenzoylmethane moiety, is observed in the complex at m/z(195, 22.47%).

A peak is seen at m/z(268, 74.54%) due to FeL<sup>++</sup>, peak due to such ion is interestingly most abundant. Quite abundant peak at m/z(592, 59.27%) observed due to oligomeric ion  $Fe_2L_2L^{++}$  similarly less intense peak noticed at m/z(698, 18.36%) due to  $Fe_2LL_2^{++}$ . The peak of the quite intensity is observed at m/z(469, 49.63%) due to  $FeL_3^{++}$ .



#### Molar Conductance

Here we measured the conductance with an alternating electrical current (I) of frequency 1 KHz and measuring the resulting voltage (V) drop in the solution. As we know that the conductance measurements can only be performed after calibration, so that cell constant value can be used for the calculating of the conductance K= $\kappa$ .G (Where  $\kappa$  = cell constant and G = conductance) precisely. Therefore, before performing the measurements on sample, the cell constant is determined with a calibration solution (0.1 N KCI). After calibration of the instrument, we have performed the measurements on our sample and the observations of conductance are shown in figure 4.

For conductance measurement we made mM solutions of  $[Fe(2-hap)_2(dbzm)]$  dissolved in DMSO. The observed Molar conductance of  $10^{-3}$ M solutions of the of  $[Fe(2-hap)_2(dbzm)]$ , lies in the range of 11 to 32 mho cm<sup>2</sup>mol<sup>-1</sup>, which is supporting their non-electrolytic behavior (see figure 4). Generally the conductance of complexes is given by modified Onsager law.



Figure 4. Room temperature Molar conductance of [Fe(2-hap)<sub>2</sub>(dbzm)] at different molar concentration in DMSO.

Figure 5. The temperature dependence of the Molar conductance of the [Fe(2hap)<sub>2</sub>(dbzm)] in DMSO.

The conductance K, is plotted against  $C^{1/2}$  and the linear portion extrapolated to zero concentration to obtain K<sub>0</sub> intercept. And the slope of the linear portion is (A+ B $\omega$ K<sub>0</sub>) which is depends on the charges of the ions concerned and reflect directly the electrolyte solution of the concerned complex. Since the term  $\omega$  involves, ionic mobility, which necessarily depends on the type of complex ions Geary (1971). It is known that the conductance also temperature dependent, and generally if the temperature increases, conductance also increases. The temperature dependence was performed at concentration 0.53985 mM of [Fe(2-hap)<sub>2</sub>(dbzm)] in DMSO. The temperature dependence of molar conductance is shown in figure 5. We can inferred from figure 5 that as the temperature is increases from 17°C to 44°C the conductance varies from 10 to 19 mho cm<sup>2</sup>mol<sup>-1</sup>. It is also observed that temperature dependence was linear in temperature range of the experiment.

#### APPLICATION

#### Antibacterial screening

Two of the ligands namely 2-hydroxyacetophenone and 1,3-diphenylpropane-1,3-dione and one of the synthesized mixed ligand complex [Fe(2-hap)<sub>2</sub>(dbzm)] were screened against the bacterial strains. The antimicrobial screening data shows (table III) that both ligands,

2-hydroxyacetophenone and 1,3-diphenylpropane-1,3-dione do not show activity against both the strains of bacteria.

It is important to note that the metal chelate exhibits more inhibitory effects towards both of the bacteria than the parent ligands. From Table 3, it is clear that the zone of inhibition is much larger for metal complexes against both bacterial strains than the parent ligands. Moreover, the complex is much powerful bactericides against *Staphylococcus aureus* and *Escherichia coli*.

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Figure 6. Antibacterial activity of [Fe(2-hap)<sub>2</sub>(dbzm)] (a) Bacterial activity against gram positive bacteria (*Staphylococcus aureus*), (b) Bacterial activity against gram negative bacteria (*Escherichia coli*)

Test compound		E. Coli Test zone of	S. Aureus Test zone of
		inhibition (mm)	inhibition (mm)
2-hydroxyacetophen	one (hap)	No activity	No activity
1,3-diphenyl propane1,3-		No activity	No activity
dione			
[Fe(2-hap) <sub>2</sub> (dbzm)]	20 ml	40	20
(10mg/ml DMSO)	40 ml	10	2
	60 ml	3	16
	80 ml	2	04
Ciprofloxacin		20	20

Table 3. Antibacterial activities of the ligands and iron metal complexes.

In our complex bacterial activity against *E.coli* bacteria is 40 mm (at 20 ml concentration) which is quite large inhibition zone then antibiotic ciprofloxacin. So our new synthesized complex can be applied safely in the treatment of urinary tract infections and some common diseases (cholecystis, cholangitis, bacteremia, neonatal meningitis, and pneumoniawhich are caused due to *E.coli* bacteria.

The increased activities of the metal chelate as compared to ligands can be explained on the basis of chelation theory. According to chelation theorySengupta et al. (1998)chelation tends to make the ligand act as more powerful and potent bactericidal agents, thus killing more of the bacteria than the ligand. It is observed that in a complex, the positive charge of the metal is partially shared with the donor atoms present in the ligands, and there may be  $\pi$ -electron delocalization over the whole chelating ring. This increases the lipophilic character of the metal chelate and favors its permeation through the lipid layer of the bacterial membranes and blocks the metal bonding sites on the enzymes of micro-organism. These complexes also disturb the respiratory processes of the cell and thus block the synthesis of protein, which restricts further growth of the organism.

There are other factors which also increase the activity, solubility, conductivity, and bond length between the metal and the ligand. Moreover, Tweedy's (1964) overtone's concept of cell permeability is also important in this contrast. According to this concept, the lipid membrane that surrounds the cell, favors the passage of only lipid-soluble material, due to which lyposolubility is also an important factor that controls the antibacterial activity of the compound.

#### CONCLUSIONS

According to the above discussion, an octahedral environment around the metal has been proposed on the basis of UV-visible and magnetic moment data. Non-electrolytic behavior has been assessed from their low conductance and high molecular weight data. All complexes are high spin paramagnetic in nature, is assessed from magnetic moment data. In the IR spectra of the complexes, shifting of v(C=O) to lower wave number side supports the chelation of the ligand to the metal atom and also support the absence of coordinated water molecules in the complexes. Mass spectral study further confirms the proposed structure of the complexes. The complexes are biologically active and exhibit enhanced antibacterial activities as compared to their parent ligands, hence further study of these complexes could lead to interesting results.

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