



NOVEL SYNTHESIS OF HETEROCYCLIC 1,3,4 THIADIAZOLE BASED BIOACTIVE METAL COMPLEXES

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Abstract: Novel Heterocyclic 1,3,4 Thiadiazole Fe(III) Complexes were prepared. Metal Complexes were prepared from ligand 4-bromo-2-(((5-mercapto-1,3,4-thiadiazol-2-yl)imino)methyl)-6-methoxyphenol with Fe(NO₃)₃.9H₂O (Iron Nitrate). Structures of complexes were confirmed based on different spectroscopic techniques like elemental analysis, FT-IR, UV-Vis, magnetic and molar conductivity measurements. All complexes were non-electrolytes and had octahedral geometry. Investigation of bioactivity for complexes and ligands exposed their moderate antibacterial activities tested *in vitro* against bacterial Strains *S. aureus* and *B. subtilis* and fungal strains of *F. Oxysporum* and *A. Niger* using Kirby-Bauer disc diffusion method.

Keywords: Heterocyclic ligand, 1,3,4 Thiadiazole, Bioactive, Octahedral.

Introduction:

There has been substantial increase in the aspire towards the discovery of effective and safe therapeutic agents for the novel drug designing due to the studies of Schiff bases and their complexes having a diverse spectrum of biological and pharmaceutical activities as antifungal^{i-iv}, antioxidant, antibacterial^{v-vii} and antitumor agents^{viii}, due to their interaction to the specific sites of a DNA-strand as reactive models for protein-nucleic acid interaction. The heterocyclic class of chelating ligands containing N, O, and S exhibits a great variety of biological activities. These activities are reported to include pharmacological applications such as antitubercular^{ix}, anticancer^x, antibacterial^{xi}, antiviral^{xii}, antifungal^{xiii}, antiinflammatory^{xiv}, antifeedant^{xv}, analgesic^{xvi}, antimicrobial^{xvii}, antimalarial^{xviii}, and anticonvulsant properties^{xix}. Azomethine group is the common structural feature of Schiff bases, where substituents may be alkyl, cycloalkyl, aryl or heterocyclic groups^{xx-xxi}. The importance of metal complexes of Schiff bases has been acknowledged in the field of bioinorganic chemistry, supramolecular chemistry, biomedical applications and material sciences^{xxii}, Metal complexes of Schiff bases provide compounds related to natural and

synthetic carriers of oxygen^{xxiii}, also provide compounds which act as active stereospecific catalysts in reduction, oxidation, hydrolysis and transformation reactions in inorganic and organic chemistry^{xxiv}, the chemistry and the applications of these novel Schiff bases thiadiazoles group containing moieties derivatives could be extensively studied by coordinating to various metal ions. As a result, the structural activity relationship study of 1,3,4-thiadiazoles could be enlarge in the future development [xxv-xxvi], Metal-binding with a ligand usually occurs using a heteroatom like nitrogen, sulphur and oxygen. Coordination bond among metal and ligand is formed, resulting in the formation of a coordination complex. Metals, as well as their complexes, have significant applications in medical history since 5000 years^{xxvii}.

In the Present study we synthesized and characterized novel Fe(III) Metal Complexes from heterocyclic ligand 4-bromo-2-(((5-mercapto-1,3,4-thiadiazol-2-yl)imino)methyl)-6-methoxyphenol. Moreover, the preliminary *in vitro* antibacterial and antifungal screening activities of the ligand are carried out and the results are reported herein.

Materials And Methods:

Experimental:

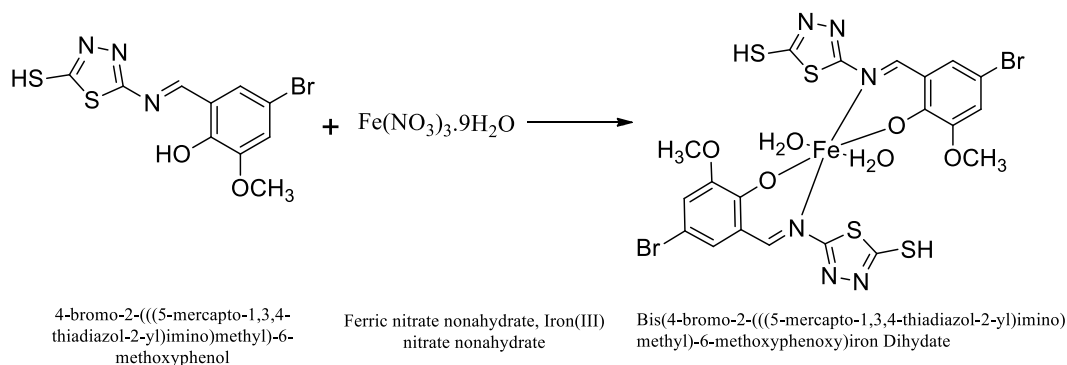
All chemical of analytical grade. All salts are metal nitrates i.e., Fe(NO₃)₃.9H₂O (Sigma-Aldrich), 5-amino-1,3,4-thiadiazole-2-thiol and 5-bromo-2-hydroxy-3-methoxybenzaldehyde from Sigma-Aldrich used without further purification. Dist. Ethanol used for synthesis of ligand, diethyl ether (Sigma-Aldrich). IR Spectra recorded on Perkin Elmer Spectrometer in range 4000-400 cm⁻¹ KBr pellets. The C, H and N analyses were carried out using a Euro-E 3000. Magnetic susceptibility measurements for the synthesized complexes were obtained at room temperature using Room Temperature magnetic moments by Guoy's method in B.M. Electronic Spectra using DMSO on Varian Carry 5000 Spectrometer.

Biological Activity:

Metal complexes (MC) of Fe (III) Was studied for their antibacterial activity against two bacteria, viz. *B. Subtilis*; *S. aureus*, Two fungi strains *A. niger* and *F. oxysporum* by Kirby-Bauer disc method^{xxviii}. The standard for antibacterial activity is Ciprofloxacin and antifungal activity is Miconazole was also screen under same condition for comparison of bioactivity.

Synthesis of Schiff base Ligand: The ligand is synthesized by reported method^{xxix}. The mixture of 1:1 5-bromo-2-hydroxy-3-methoxybenzaldehyde (2.31g, 0.01mol) with 5-amino-1,3,4-thiadiazole-2-thiol (1.33 g, 0.01 mol) dissolved in ethanol. Then add Few drops of glacial acetic acid was added. The resultant mixture stirred for 4-5 hrs the yellowish colored precipitate of Ligands was obtained. Wash with Ethanol recrystallised with Ethanol and Ether then dried. The purity of compound was checked by TLC using Silica Gel method.

Synthesis of Metal Complexes: The metal complexes were prepared by mixing of metal nitrates i.e. Fe(NO₃)₃.9H₂O with (30 ml) ethanolic solution of Ligand 4-bromo-2-(((5-mercapto-1,3,4-thiadiazol-2-yl)imino)methyl)-6-methoxyphenol in (metal: ligand) 1:2 ratio. The resulting mixture refluxed on water bath for 5-6hr. A colored product obtain washed with ethanol, filtered, and recrystallised with ethanol (Scheme.1)



Scheme 1. Synthesis of metal complexes

Results and Discussion:

The ligand 4-bromo-2-(((5-mercapto-1,3,4-thiadiazol-2-yl)imino)methyl)-6-methoxyphenol and its Metal Complexes of Fe(III) is stable at room temperature in solid state (Scheme 1). The ligand and its Metal Complexes is soluble in organic solvent like DMSO, DMF. The physical and analytical data shown in Table 1. Spectral evaluation shows formation of ligand and its metal complexes. The synthesized complexes having 1:2 metal to ligand stoichiometric ratio.

Table 1: Proposed Structures of metal complexes M: Fe (III).

Entry	Products	Time (h)	Yield (%)	M.P (°C)
MC ₁		5-7	72	>300

Characterization data of Heterocyclic Ligands:

4-bromo-2-(((5-mercapto-1,3,4-thiadiazol-2-yl)imino)methyl)-6-methoxyphenol

(Table.1,L) : Dark Yellow; M.F. $C_{10}H_8BrN_3O_2S_2$; Yield : 72% ; M.P. 230°C; Molar Cond. (DMSO 1×10^{-3} conc., $\text{ohm}^{-1} \text{cm}^2 \text{mol}^{-1}$): 6.62; UV (DMSO, cm^{-1}): 282 ($\pi \rightarrow \pi^*$ tran. of benzene ring), 368 ($n \rightarrow \pi^*$ azomethine moieties and phenolic -OH.); IR (KBr cm^{-1}): $\nu = 3323$ (O-H str. in aromatic ring), $\nu = 1633$ (C=N azomethine), $\nu = 1493$ (-C=N-N=C str. in Thiadiazole ring), $\nu = 1268$ (C-O Phenolic), $\nu = 1026$ (N-N Thiadiazole ring), $\nu = 756$ (C-S-C str. in thiadiazole ring); $^1\text{H NMR}$ (400 MHz, DMSO- d_6) δ ppm: $\delta = 11.23$ (s, 1H, Ar-OH), $\delta = 8.80$ (s, 1H, CH=N), $\delta = 7.10-7.56$ (s, 2H, Ar-CH); $^{13}\text{C NMR}$ (DMSO- d_6 , 400 MHz,) δ ppm: $\delta = 150.6-121.6$ (C_1-C_6 Aromatic), $\delta = 161.7$ (C_7 , -C=N- Azomethine), $\delta = 155.6$ (C_8 Thiadiazole ring), $\delta = 181.3$ (C_9 Thiadiazole ring), $\delta = 52.2$ (C_{10} -OCH₃-); MS (70 eV) m/z : 347 [M+H, 100%], Anal. Calcd. For $C_{10}H_8BrN_3O_2S_2$: C, 34.69; H, 2.33; N, 12.14; S, 18.52. Found: C, 34.60; H, 2.31; N, 12.02; S, 18.46.

Characterization of Heterocyclic Metal Complexes:

Bis(4-bromo-2-(((5-mercapto-1,3,4-thiadiazol-2-yl)imino)methyl)-6-methoxyphenoxy)iron Dihydrate

Light Green; M.F. $C_{20}H_{18}Br_2FeN_6O_6S_4$; Yield : 72% ; M.P. >300°C; Molar Cond. (DMSO 1×10^{-3} conc., $\text{ohm}^{-1} \text{cm}^2 \text{mol}^{-1}$): 11.8; Magnetic moment

(μ_{eff} B.M.) :5.89;UV(DMSO, cm^{-1}) :392,460,585 ($\pi \rightarrow \pi^*$, $n \rightarrow \pi^*$, d-d transition); IR(KBr Cm^{-1}) : $\nu = 3446$ (O-H str. Water molecule), $\nu = 1620$ (C=N azomethine), $\nu = 1469$ (-C=N-N=C (str.in Thiadiazole ring), $\nu = 1290$ (C-O Phenolic), $\nu = 1020$ (N-N Thiadiazole ring), $\nu = 751$ (C-S-C str.in thiadiazole ring), $\nu = 554$ (M-O bond), $\nu = 442$ (M-N bond); Anal.Calcd.For $\text{C}_{20}\text{H}_{18}\text{Br}_2\text{FeN}_6\text{O}_6\text{S}_4$: C, 30.71; H, 2.32; N, 10.74; Fe, 7.14; Found : C, 30.15; H, 2.02; N, 10.20; Fe, 7.01;%.

Results and Discussion:

Infrared spectra has proven to be the most suitable technique to give enough information to elucidate the nature of bonding of the ligand to the metal ions. The IR data the spectra of Bis(4-bromo-2-(((5-mercapto-1,3,4-thiadiazol-2-yl)imino)methyl)-6-methoxyphenoxy)iron Dihydrate heterocyclic metal complexes show peak at 3446 cm^{-1} of O-H stretching of Water molecule while in free ligand 3323 cm^{-1} . Difference of -OH Stretching frequency in ligand and metal complexes is due to water molecule coordinate to metal in metal complexes^{xxx}. The azomethine peak of ligand observed at 1633 cm^{-1} (-C=N- azomethine) while in metal complexes it is observed at 1620 cm^{-1} . When electron donation from ligand azomethine nitrogen to metal takes place in metal complexes the stretching frequency of MC decreases due to chelation it confirms metal to ligand bonding through azomethine nitrogen^{xxxii}. The phenolic (C-O) stretching frequency of ligands and MC is 1268 cm^{-1} & 1290 cm^{-1} respectively. The difference is due to electron donation of oxygen to metal in MC, it also shows band M-O at 554 cm^{-1} . The coordination of azomethine nitrogen also confirms when we see the band M-N at 442 cm^{-1} in Metal complexes^{xxxiii}.

The band is at 282nm and 398 nm in ligand is due to transition in benzene ring of ligand and phenolic OH and Azomethine moieties. These bands shift to longer wavelength due to formation for Schiff base metal complexes at 392,460,585 having $\pi \rightarrow \pi^*$, $n \rightarrow \pi^*$, d-d transition^{xxxiii}. Molar Conductance value of MC is 11.8 shows non-electrolytic nature of metal complexes. The magnetic moment value for Fe (III) complexes (MC) is 5.89 B.M hence it confirms octahedral geometry of metal complexes^{xxxiv}.

Antimicrobial Activity

The antimicrobial activity of heterocyclic metal complexes were studied against two gram positive bacteria *S. aureus* and *B. Subtilis* two fungi *A. niger* and *F. Oxysporum*. heterocyclic metal complexes show better antimicrobial activity as compared to ligand. The Antifungal and Antibacterial activity of ligand and MC shown in table 2. Metal complexes show more activity due to chelation it shows delocalization of π electron on chelating ring it enhances penetration of complexes in lipid membrane of microorganism it blocks the binding site enzymes of harmful microorganism some factors like hydrophilicity lipophilicity, solubility, Conductivity & bonding between M-L that increases the activity of metal complexes. lipophilic nature of the complexes arising due to chelation. It is probably due to faster diffusion of the chelates as a whole through the cell membrane or due to the chelation theory^{xxxv-xxxvii}.

Table 2. Antimicrobial activity of ligand and its Metal Complexes

Compounds	Antibacterial Activity		Antifungal Activity	
	<i>S.aureus</i>	<i>B.subtilis</i>	<i>A.niger</i>	<i>F.oxysporum</i>
	Diameter of inhibition Zone in mm	Diameter of inhibition Zone in mm	Diameter of inhibition Zone in mm	Diameter of inhibition Zone in mm
	500ppm	500ppm	500ppm	500ppm
Ligands(HL)	18	18	14	22
Fe Complex	23	23	20	21
Ciprofloxacin(Standard)	34	33	---	---
Miconazole(Standard)	---	---	31	27

Conclusion

In the Present Study new Heterocyclic 1,3,4 Thiadiazole based bioactive Metal complexes (MC) i.e. Bis(4-bromo-2-(((5-mercapto-1,3,4-thiadiazol-2-yl)imino)methyl)-6-methoxyphenoxy)iron Dihydrate was prepared by Conventional method. The metal complex is more Bioactive as compare to parent ligand against all pathogenic microorganism. These type of study help to overcome problems like multi-drug resistance of microorganism.

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