Available Online at

http://www.ijcpa.in

July-September 2018

International Journal of
CHEMICAL AND PHARMACEUTICAL ANALYSIS

eISSN: 2348-0726 ; pISSN : 2395-2466

Research Article | Volume-5 | Issue-4 | Article ID: 1449

BIOLOGICAL SCREENING OF NEWLY SYNTHESIZED SCHIFF BASE, (Z)-2-(1-(2, 5-DIMETHOXYPHENYLIMINO) ETHYL) PHENOL AND ITS COMPLEXES

R. Sathya* and A. Xavier

PG & Research Department of Chemistry, The Madura College, Madurai-625011, Tamil Nadu, India.

Corresponding Author: *Email: sathyasriramr13@gmail.com*

Received: 19 February 2018 / Revised: 13 August 2018 / Accepted: 25 August 2018 / Available online 30 September 2018

ABSTRACT

Some new mixed metal complexes were synthesized using (Z)-2-(1-(2,5-dimethoxyphenylimino) ethyl) phenol ligand (L) and metal ions (M) = Cu (II), Mn (II), V (II) and Au (III). The ligand and metal complexes were characterized quantitatively and qualitatively by using, Elemental analysis, Infrared spectra, Electronic spectra. Elemental analysis of the metal complexes was suggested that the stoichiometry ratio is 1:2 (metal-ligand). Fluorescent studies also have been reported. The electronic spectrum provides clear idea about the metal complexes. The Schiff base complexes have been screened for their invitro biological activities against and bacteria and fungi. Anti-bacterial activity against three bacteria via; [Staphylococcus aureus, Bacillus sp., **(***Gram positive) Pseudomonas aeruginosa (Gram negative)] and Antifungal activities (Aspergillus Niger and Aspergillus flavus). Anti-oxidant properties of metal complexes also have been studied. The metal complexes show more potential activities compared with Schiff base ligands.*

Keywords – *Schiff base, Transition metal complexes, spectroscopy, Fluorescent studies, Biological Screening, Anti-oxidant studies.*

1. INTRODUCTION

The Schiff bases are widely employed as ligands in complex formation. They coordinate with many transition metal ions producing metal complexes that display motivating physical, chemical, biological and catalytic properties [1-8]. Schiff bases, which are condensation products of primary amines and aldehydes or ketones. Analysis on metal-organic complexes represents one of the most dynamic areas of material science and chemical research due to their stimulating properties and potential in various applications as host–guest chemistry, ion exchange and catalysis.

Transition metal complexes have extensive applications in food industry, dye industry, analytical chemistry, catalysis, agrochemical and biological activities [9]. Novel dyes especially those used on some of the artificial fabrics that are difficult to tint, have been prepared from coordination compounds. Chelated iron is used in fertilizers to provide iron. Research has shown significant progress in utilization of transition metal complexes as drugs to treat several human diseases like carcinomas, lymphomas, infection control, anti-inflammatory, diabetes, malaria and neurological disorders [10].

Schiff bases are also used as optical and electrical sensors as well as in various chromatographic methods, to enhance selectivity and sensitivity [11]. Schiff bases have likewise structural resemblance with natural biological systems [12,13]. Many biologically vital Schiff bases exhibiting, antimicrobial, antibacterial, antifungal, anti-inflammatory, antiplatelet and antitumoral agents are reported in the literature [14-16].

The biological activity of the metal complexes is focused by the following factors (i) the chelate effect of the ligands, (ii) the nature of the donor atoms, (iii) the total charge on the complex ion, (iv) the nature of the metal ion, (v) the nature of the counter ions that neutralize the complex, and (vi) the geometrical structure of the complex [17]. Besides, chelation diminishes the polarity of the metal ion because of partial sharing of its positive charge with the donor groups and possibly the p-electron delocalization within the whole chelate ring system that is formed during coordination [18].

These factors increase the lyophilic nature of the central metal atom and hence increasing the hydrophobic character and liposolubility of the molecule favoring its permeation through the lipid layer of the bacterial membrane. This enhances the rate of uptake/entrance and thus the antibacterial activity of the testing compounds. The Schiff base ligand and its complexes were tested for their inhibitory effects on the growth of bacteria: Staphylococcus aureus (G+), Pseudomonas aeruginosa and Bacillus species and fungi *Aspergillus niger* and *Aspergillus flavus.*

Over production of activated oxygen species in the forms of superoxide anion (O_2^-) and hydroxyl radical ($HO^·$) is considered to be the main contributor to oxidative damage of biomolecules such as DNA, lipids and proteins, thus accelerating cancer, aging inflammation, cardiovascular and neurodegenerative ailments [19]. Many transition metal complexes also revealed interesting antioxidant activities [20].

We have synthesized and characterized a novel Schiff base ligand (Z)-2-(1-(2, 5-dimethoxyphenylimino) ethyl) phenol and its Cu (II), Mn (II), V (II) and Au (III) complexes. They were characterized by elemental analyses, UV, FT-IR, Fluorescent studies. The antimicrobial activity and anti-oxidant activity of these compounds were examined systematically.

2. MATERIALS AND METHODS

Reagents

All the chemicals and solvents were of AR grade. All the reagents used for the preparation of the ligand and metal complexes were obtained from Sigma Aldrich.

Apparatus

The electronic spectra of the ligand and their complexes have been recorded Schimadzu UV-1800 in DMSO solvent in the range of 200—800nm. FT-IR spectra recorded using KBr pellets in Schimadzu FT-IR 8201 spectrometer (4000-400 cm^{-1}).

Synthesis of Ligand (L)

The (Z)-2-(1-(2,5-dimethoxyphenylimino)ethyl)phenol ligand (L) was prepared by the following method: 2'-hydroxyacetophenone (20m.mol) was added to 2,5-dimethoxy aniline (20 m.mol) in ethanol was stirred in a round bottom flask . The reaction mixture was stirred and refluxed for 6 hrs. The progress of the reaction was monitored by TLC. The resulting precipitate was filtered and washed and dried in an air oven.

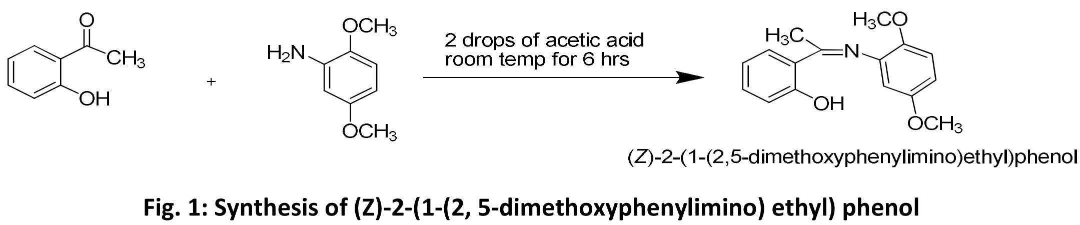
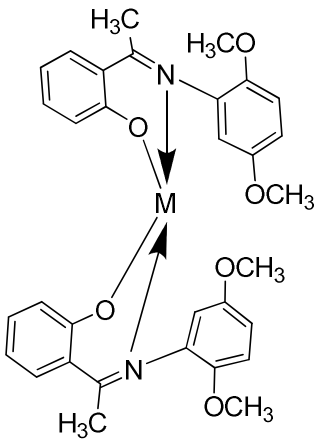


Fig. 1: Synthesis of (Z)-2-(1-(2, 5-dimethoxyphenylimino) ethyl) phenol

2.2. Synthesis of Schiff Base metal Complexes

Cu(II), Mn(II),V(II) and Au(III) complexes were prepared by using the following general procedure: Metal salt (10m.mol) was added to a ethanolic solution of the ligand followed by refluxing for 6 hours. The volume of the obtained solution was reduced to one-half its original volume by evaporation. The precipitated product was then filtered and finally dried under vacuum. The purity of the synthesized complexes was checked by TLC.



Where M = Cu (II), Mn (II), V (II) and Au (II)

Fig. 2: Proposed structure of Metal Complex

2.3. Spectral Characterization

UV-Visible Spectra

The UV-visible electronic spectra (200-800 nm) were recorded by UV-Vis-1800 series (Shimadzu) double beam spectrophotometer using DMSO solvent.

This measurement gives the information about the stereochemistry of metal ions in the complexes. It is very beneficial to measure the number of conjugated double bonds and also aromatic conjugation within the various molecules. The electronic absorption spectra of the Schiff base and its metal complexes were recorded at room temperature.

FT-IR spectra

FT-IR spectra were recorded using KBr pellets in Schimadzu FT-IR 8201 spectrometer (4000 – 400 cm^{-1}).

2.4. Fluorescent Studies

Fluorescent transition metal centers are particularly attractive moieties because they often possess distinctive electrochemical or photo physical properties thus enhancing the functionality of the binding agent. The fluorescence characteristics of metal complexes were studied at room temperature in solid state.

2.5. Biological Activity

Antimicrobial activity and Antifungal activity of ligand and metal complexes

Antibacterial activity of the ligand and metal complexes was determined using well diffusion method. It was performed by sterilizing Muller Hinton agar media. After solidification, wells were cut on the Muller Hinton agar using cork borer. The test bacterial pathogens were swabbed onto the surface of Muller Hinton agar plates. Wells were impregnated with 25µl of the test samples. The plates were incubated for 30 min to allow the extract to diffuse into the medium. The plates were incubated at 30°C for 24 hrs. and then the diameters of the zone of inhibition were measured in millimeters. Each antibacterial assay was performed in triplicate and mean values were reported.

Similarly, Antifungal activity of the ligand and metal complexes was determined using well diffusion method. Each antifungal assay was performed in triplicate and mean values were reported.

2.6. Anti-oxidant assay

The radical scavenging activity of different compounds was determined by using DPPH assay according to Chang et al. (2001). The decrease in the absorption of the DPPH solution after the addition of an antioxidant was measured at 517nm. Ascorbic acid (10mg/ml DMSO) was used as reference. (DPPH)1, 1 Diphenyl 2- Picryl Hydrazyl is a stable (in powder form) free radical with red color which turns yellow when scavenged. The DPPH assay uses this character to show free radical scavenging activity. The scavenging reaction between (DPPH) and an anti-oxidant (HA) can be written as,

$$(DPPH) + (H\text{-}A) \longrightarrow DPPH\text{-}H + (A) \quad \text{-------------------------------------}> 1$$

DPPH radical was scavenged by antioxidants through the donation of hydrogen, forming the reduced DPPH-H radical. The colour changed from purple to yellow after reduction.

Anti-oxidants react with DPPH and reduce it to DPPH-H and as consequence the absorbance decreases. The degree of discoloration indicates the scavenging potential of the antioxidant compounds in terms of hydrogen donating ability. Different volumes of test solutions were made up to 40µl with DMSO and 2.96ml DPPH (0.1mM) solution was added. The reaction mixture was incubated in dark condition at room temperature for 20 min. After 20 min, the absorbance of the mixture was read at 517 nm. DPPH was taken as control. The % radical scavenging activity of the complexes was calculated using the formula,

$$\% \text{ RSA} = \text{Abs. control x Abs. sample} \,/\, \text{Abs. control} \quad \text{-----------------------------}> 2$$

Where, RSA is the Radical Scavenging Activity, Abs control is the absorbance of DPPH radical + ethanol, Abs sample is the absorbance of DPPH radical + complex.

A calibration curve was plotted with % DPPH scavenged vs. concentration of standard anti-oxidant. A percent inhibition versus concentration curve was plotted and the concentration of sample required for 50% inhibition was determined and expressed as IC_{50} value. The lower the IC_{50} value indicates high antioxidant capacity.

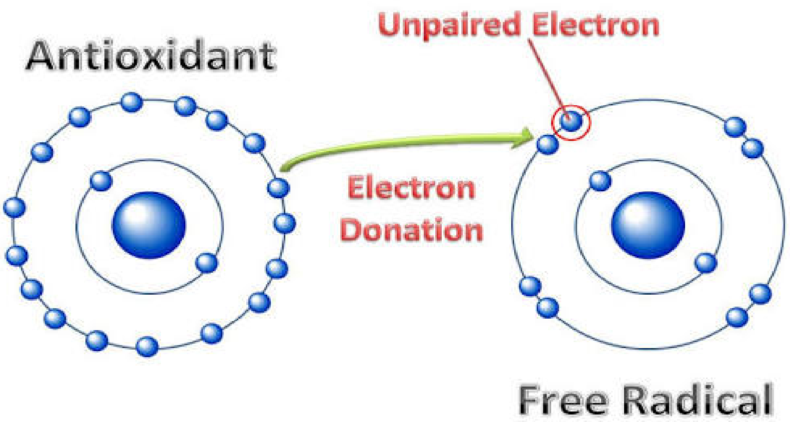


Fig. 3: Electron donation from Anti-oxidant to free radical

3. RESULTS AND DISCUSSION

3.1. Elemental Analysis

The micro-elemental analysis for C, H, N and O as well as the molecular weight of the complexes obtained were in agreement with the predicted formula for complexes.

Table 1: Analytical and Physical data of ligand and its complexes

Compounds	Physical appearance	Melting point (°C)	Elemental (%) Calc. (found)					m/z
			C	H	N	O	M (metal)	
Ligand (L_1)	Brown	84	70.83 (70.85)	6.32 (6.31)	5.16 (5.15)	17.69 (17.67)	–	271.12
Cu – L	Black	118	63.62 (63.60)	5.34 (5.35)	4.64 (4.65)	15.89 (15.91)	10.52 (10.50)	603.16
Mn – L	Pink	125	64.54 (64.55)	5.42 (5.40)	4.70 (4.72)	16.12 (16.11)	9.22 (9.20)	595.16
V – L	Brown	129	64.97 (64.99)	5.45 (5.42)	4.74 (4.76)	16.23 (16.20)	8.61 (8.63)	591.17
Au – L	Brown	140	52.11 (52.14)	4.37 (4.39)	3.80 (3.78)	13.02 (13.04)	26.70 (26.73)	737.19

3.2. FT-IR spectra

The IR spectra of the ligand showed a broad band of medium intensity at 3200 cm^{-1} for ligand characteristic of H-bonded (O–H) of the phenolic groups. Comparison of the IR spectra of the metal complexes with those of the studied ligand showed that all complexes have a broad medium or strong band in the range 3384-3500 cm^{-1} attributable to (OH) of the coordinated water molecule in the complexes. The medium intense band observed at 1273 cm−1 for ligand was ascribed to the phenolic C–O stretching mode.

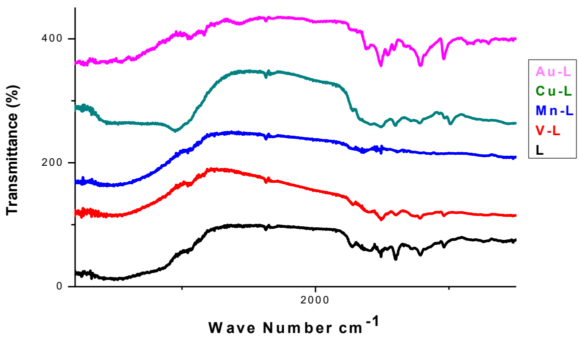


Fig. 4: FT-IR spectra of Metal Complexes.

3.3. Fluorescent studies

The Fluorescence characteristics of ligand and metal complexes were studied at room temperature in solid state. Fluorescence spectra of complexes show red shift, which may be due to the chelation by the ligands to the metal ion. It enhances the ligand ability to accept electrons and decreases electron transition energy. In metal complexes M→L co-ordination may lead to significant changes of the fluorescence properties of the ligand, including increase or decrease of the intensity.

The emission spectra of ligand and Cu(II), Mn(II), V(II), Au(III) complexes are characterized by an emission band around Cu = 345nm & 589nm, Mn = 540nm, V = 356nm &374nm& 566nm, Au = 354nm & 598 nm in DMSO. The Ligand exhibits an excitation maximum wavelength at 350 nm which creases and shifts to region. This shows that ligand and complexes are having fluorescent character.

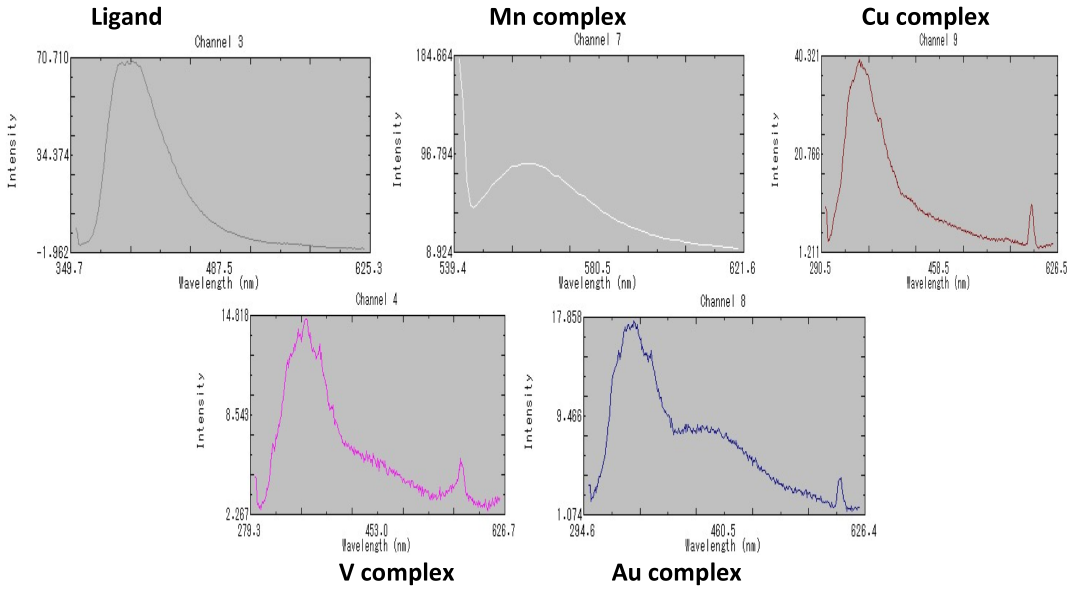


Fig. 5: Fluorescent spectra of Ligand and Metal Complexes.

3.4. Antibacterial activity of ligand and complexes

The antibacterial activity for ligand and complexes were listed in Table 2. The results indicated that, Cu(II) complex showed high activity whereas Mn(II) and V(II) complexes showed moderate activity against all three bacteria. Au (III) complex showed low activity. The different complexes exhibited weak to high activity against the bacterial strains when compared with ligand. The bactericidal activity of the complexes follow the order, Cu (II) > V(II) > Mn(II) >Au(III). The potential activity of copper complex than other metal complexes is presumed from the fact that the polarization decreases with the increase in the size of the metal ion [21] . The antibacterial activity exhibited by the metal complexes is comparatively greater than the corresponding ligands.

Table 2: Antibacterial activity of the ligand and complexes

Test organisms	Zone of inhibition in millimeter (in diameter)						
	Ligand	Mn	Cu	Au	V	Solvent control	Standard Amikacin 30µg
Pseudomonas aeruginosa	20	21	23	18	22	NZ	25
Staphylococcus aureus	24	20	26	22	25	NZ	27
Bacillus sp.,	23	25	26	20	24	NZ	28

Solvent used: DMSO (Dimethyl Sulphoxide) **Standard used**: Amikacin 30 µg. **C**- Control.

Plates: 1, 2, 3: Antibacterial activity of the Ligand and Cu, Mn,V and Au Complexes treated against *Pseudomonas aeruginosa.*

Plates: 4, 5, 6: Antibacterial activity of the Ligand and Cu, Mn,V and Au Complexes treated against *Staphylococcus aureus.*

Plates: 7, 8, 9: Antibacterial activity of the Ligand and Cu, Mn, V and Au Complexes treated against *Bacillus species.*

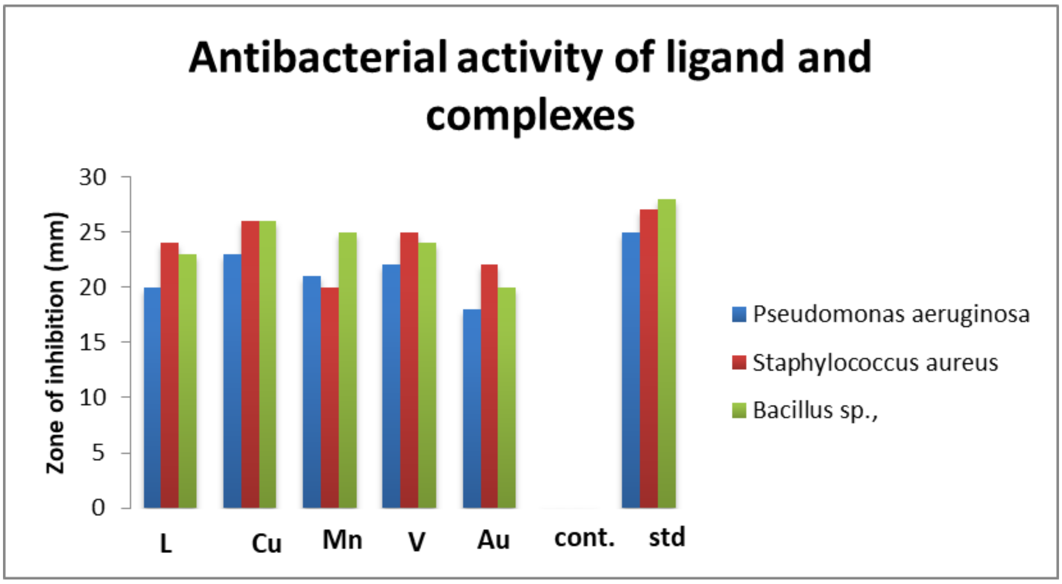


Fig. 6: Antibacterial activity of Ligand and Metal Complexes.

3.5. Antifungal activity of Ligand and complexes

From the results it is inferred that the Schiff base complexes have more antifungal activity when compared to the ligand. Among the complexes Mn (II) and Cu (II) complexes have more activity.

Table 3: Antifungal activity of the ligand and complexes

Test Fungal Pathogens	Zone of inhibition in millimeter (in diameter)						
	Ligand	Mn	Cu	Au	V	Solvent control	Standard Cotrimoxazole 25 µg
Aspergillus niger	19	25	23	17	20	NZ	27
Aspergillus flavus	21	23	24	19	17	NZ	29

Solvent used: DMSO (Dimethyl Sulphoxide) **Standard used**: Cotrimoxazole 25 µg. **C-**Control

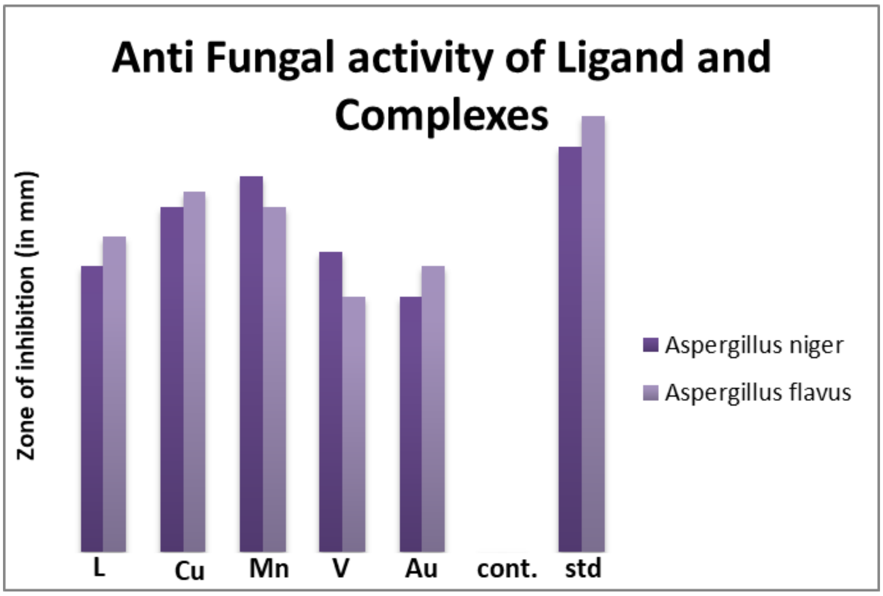
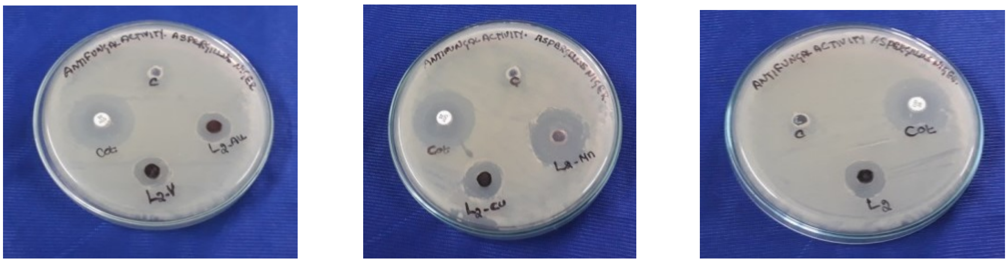
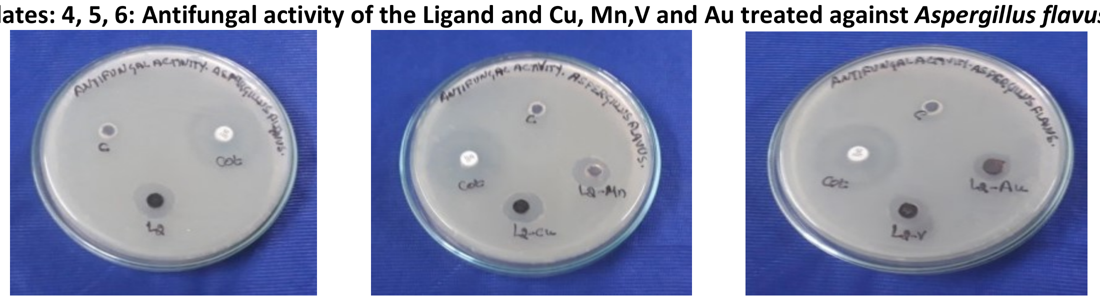


Fig. 7: Antifungal activity of Ligand and Metal Complexes.



Plates: 1, 2, 3: Antifungal activity of the Ligand and Cu, Mn,V and Au Complexes treated against *Aspergillus Niger.*

Plates: 4, 5, 6: Antifungal activity of the Ligand and Cu, Mn,V and Au treated against *Aspergillus flavus.*



3.6. Anti-oxidant Property

The human body has enzymatic and non-enzymatic antioxidant protections which hinder the harmful effect of free radicals and other oxidants. A large number of diseases including cancer cardiovascular diseases and neural disorders are owing to the formation of free radicals in the human body tissues. The main role of antioxidant is refining the quality of life by inhibiting the formation of free radicals in human body. In the antioxidant activity study, the DPPH assay is a potent way to predict the scavenging capability of ligand and its complexes.

The data reveal that copper complex is having good inhibitor ability than the other complexes. The complexes of Mn(II), V(II) and Au(III) have minimum inhibitor ability compared to the standard (Vitamin C). However, the complexes having enriched antioxidant activity protect the human body against various harmful diseases.

Table 4: Antioxidant Property of the Ligand and complexes

S. No	DPPH (ml)	Inhibition concentration [I]	Buffer	Absorbance			
				Cu	Mn	V	Au
1	2.5	-	7.5	0.065	0.059	0.063	0.054
2	2.5	0.002	7.1	0.057	0.051	0.055	0.049
3	2.5	0.004	6.7	0.049	0.045	0.048	0.041
4	2.5	0.006	6.3	0.040	0.037	0.039	0.036
5	2.5	0.008	5.9	0.036	0.031	0.034	0.028
6	2.5	0.010	5.5	0.021	0.015	0.019	0.013

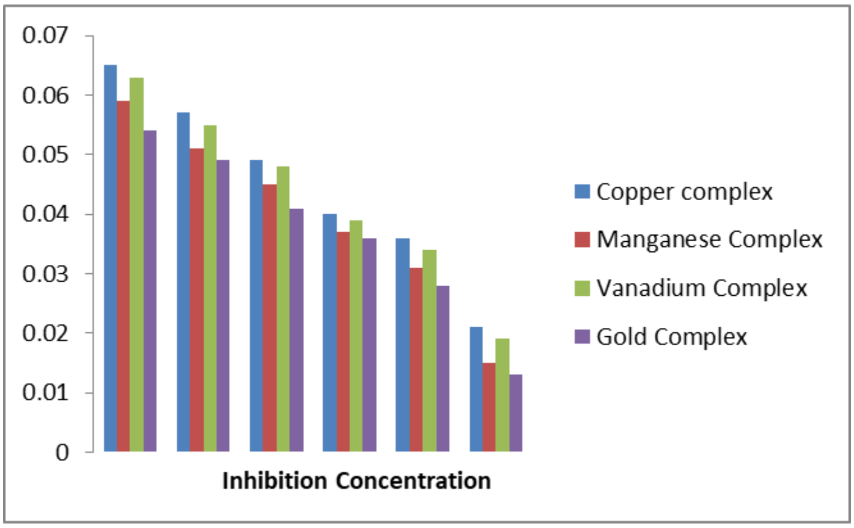


Fig. 8: Anti-oxidant activity of Metal Complexes

4. CONCLUSION

The synthesis and characterization of (Z)-2-(1-(2,5-dimethoxyphenylimino)ethyl)phenol and its Cu(II),Mn(II),V(II), Au(III) have been attained with physicochemical, spectral and antimicrobial activities. The IR, UV spectral studies lead to the conclusion that the ligand coordinated through azomethine nitrogen and ketonic oxygen atoms. From the spectral data, it is found that the geometrical structures of the complexes are octahedral. The metal complexes showed superior activity than the ligand. The copper (II) complex has the higher antimicrobial activity than the other complexes. The activity of the compounds found to be in the order of Cu (II) > V(II) > Mn(II) >Au(III). As far as our outcomes are concerned, these complexes can be used as potential chemotherapeutic agents and hold much promise in the field of drug discovery.

REFERENCES

1. LI, Jun-Ling, et al. Self-activated DNA cleavage of a water-soluble mononuclear Cu (II) complex with polyquinolinyl ligand. Journal of Coordination Chemistry, 2014, 67.22: 3598-3612.
2. Wani, Waseem A., et al. Copper (II), nickel (II), and ruthenium (III) complexes of an oxopyrrolidine-based heterocyclic ligand as anticancer agents. Journal of Coordination Chemistry, 2014, 67.12: 2110-2130.
3. Subha, L., et al. Mixed ligand Cu (II) complexes containing o-vanillin-L-tryptophan Schiff base and heterocyclic nitrogen bases: synthesis, structural characterization, and biological properties. Journal of Coordination Chemistry, 2015, 68.6: 1021-1039.
4. Thamilarasan, Vijayan; Jayamani, Arumugam; Sengottuvelan, Nallathambi. Synthesis, molecular structure, biological properties and molecular docking studies on MnII, CoII and ZnII complexes containing bipyridine–azide ligands. European journal of medicinal chemistry, 2015, 89: 266-278.

5. Shamsi, Manal; Yadav, Shipra; Arjmand, Farukh. Synthesis and characterization of new transition metal {Cu (II), Ni (II) and Co (II)} l-phenylalanine–DACH conjugate complexes: in vitro DNA binding, cleavage and molecular docking studies. Journal of Photochemistry and Photobiology B: Biology, 2014, 136: 1-11.
6. Fasina, Tolulope M.; Ogundele, Olorunfemi O.; AYENI, Isaac. Synthesis and biological properties of N2O2 Schiff bases derived from o-phenylenediamine and substituted salicylaldehydes. 2014.
7. Mohammed Fakruddin Ali Ahmed and V. Mahammad Yunus. Microwave Synthesis and Antimicrobial Activity of Some Cu (II), Co (II), Ni (II) and Cr (III) complexes with Schiff base 2,6-Pyridine dicarboxaldehyde Thiosemicabazone, Oriental Journal Of Chemistry, 2014; 30(1): 111-117.
8. P.K. Sasmal, R. Majumdar, R.R. Dighe, A.R. Chakravarty, Dalton Trans. 39 (2010) 104-113.
9. Siga, Fatma, et al. Superb efficient and recycle polymer-anchored systems for palladium catalyzed Suzuki cross-coupling reactions in water. Applied Catalysis A: General, 2012, 449: 172-182.
10. Gwaram, Nura Suleiman, et al. Antibacterial evaluation of some Schiff bases derived from 2-acetylpyridine and their metal complexes. Molecules, 2012, 17.5: 5952-5971.
11. Boruah, Jeena Jyoti, et al. Polymer-anchored peroxo compounds of molybdenum and tungsten as efficient and versatile catalysts for mild oxidative bromination. Polyhedron, 2013, 52: 246-254.
12. Sathyaraj, Gopal; Weyhermüller, Thomas; NAIR, Balachandran Unni. Synthesis, characterization and DNA binding studies of new ruthenium (II) bisterpyridine complexes. European journal of medicinal chemistry, 2010, 45.1: 284-291.
13. Kotkar, Shubhangi N.; Juneja, Harjeet D. Synthesis, characterization, and antimicrobial studies of N, O donor Schiff base polymeric complexes. Journal of Chemistry, 2013, 2013.
14. M. Aslam, I. Anis, N. Afza, A. Hussain, L. Iqbal, J. iqbal, Zaitoon Ilyas, S.Iqbal, A. H. Chaudhry and M. Niaz, International Journal of Current Pharmaceutical Research, 2012, 4(4)42-46.
15. Pope, L. H., et al. Atomic force microscopy studies of intercalation-induced changes in plasmid DNA tertiary structure. *Journal of microscopy*, 2000, 199.1: 68-78.
16. Rollas, Sevim; Küçükgüzel, S. Güniz. Biological activities of hydrazone derivatives. *Molecules*, 2007, 12.8: 1910-1939.
17. R. Sathya, A. Xavier. Int. J. Curre. Resea. 9 (2017) 62032-62039.
18. Vicini, P.; Lncertim, Doytchmnova IA; Colla, P. L. Busonera 8, Loddo RE. Synthesis and antiproliferative activity of benzo [djisothiazole hydrazones. Eur J Med Chem 2006, 41.53: 624-632.
19. Shahabadi, Nahid; Falsafi, Monireh; Moradi Fili, Soraya. Synthesis, characterization, molecular modeling, and DNA interaction studies of a Cu (II) complex containing drug of chronic hepatitis B: adefovir dipivoxil. Journal of Coordination Chemistry, 2015, 68.8: 1387-1401.
20. C. Basu, S. Chowdhury, R. Banerjee, H.S. Evans, S. Mukherjee, Polyhedron., 2007, 26, 3617-3264.
21. Ji HF, Zhang HY. A new strategy to combat Alzheimer's disease. Combining radical-scavenging potential with metal-protein-attenuating ability in one molecule. Chem Res Toxicol, 2004, 17:471–475.