Heterocyclic Letters
Vol. 15| No.4|869-878|August-October|2025
ISSN : (print) 2231–3087 / (online) 2230-9632
CODEN: HLEEAI
http://heteroletters.org

SYNTHESIS, CHARRACTERIZATION AND BIOLOGICAL INVESTIGATIONS OF SCHIFF BASE LIGANDS.

Ashish Bansod [a], Rajesh Deshmukh [b] and Anand Aswar [c]

[a] *Department of Chemistry, Rajarshee Shahu Science College Chandur Rly, 444904, India*
[b] *Department of Chemistry, R.D.I.K. College Badnera Rly Dist.Amravati, 444701, India*
[c] *Department of Chemistry, Sant Gadge Baba Amravati University, Amravati, (Maharashtra), 444602, India.*
[a] *Corresponding author: drashishbansod@gmail.com*

ABSTRACT:
Synthesis of a new Schiff base derived from 2-hydroxy-5-methylacetophenone, 2-hydroxy-5-chloro-4-methylacetophenone and 2-hydroxy-5-chloroacetophenone and glycine. The ligands have been characterized on the basis of analytical data, IR. The ligands are a dibasic tridentate (ONO) donor. Antibacterial activities of ligand have been determined by screening the compounds against various Gram (+) and Gram (−) bacterial strains.

KEYWORDS: 2-hydroxy-5-chloroacetophenone and glycine and biological investigations.

INTRODUCTION:

Schiff Base ligands played a vital role in the development of coordination chemistry [i-ii]. All kinds of natural amino acids that have been used to synthesize Schiff bases are found to be very effective metal chelators. Several amino acid and Schiff base [iii-iv] are potential models for a number of important biological systems. The present article describes the synthesis and structural, thermal, and antibacterial studies of the Schiff base derived from 2-hydroxy-5-methylacetophenone,2-hydroxy-5-chloro-4-methylacetophenone and 2-hydroxy-5-chloroacetophenone[v] and glycine.

EXPERIMENTAL:

GENERAL PROCEDURE:

The infrared spectra were recorded using KBr on a Shimadzu 8201 spectrophotometer in the range 400-4000cm^{-1}. The carbon, hydrogen, and nitrogen analyses were carried out on a Carlo Erba 1108 elemental analyzer. ^{1}H and ^{13}C NMR spectra of the ligands were recorded on Bruker Advance II, 400MHz, NMR spectrophotometer in DMSO-d_6 with TMS as an internal standard. Magnetic measurements were carried out by the Sherwood magnetic susceptibility balance MK-1 at room temperature. The chloride contents were determined as AgCl by following a standard procedure. Mass spectra were recorded on a Waters, Q-TOF micro mass (LC-MS) spectrometer. The surface morphology was observed using a JEOL Model JSM-6390LV scanning electron microscope.

Antimicrobial Activity

The Schiff base ligands were screened for their anti-bacterial and anti-fungal activity against *Escherichia Coli* (MTCC 390), *Shigellaflexneri {Mice \A51), Salmonella typhi* (MTCC 733), *Proteus vulgaris* (MTCC 4226),*Bacillus coagulans, (MTCC 2302),Pseudomonas aeruginosa* (MTCC 424),*Staphylococcus aureus* (MTCC 96),*Klebsiella pneumoniae {yiTCC* 109),*Salmonella typhimurium* (MTCC 98), *Enterococcus faecalis* (MTCC 439), *Staphylococcus epidermidis* (MTCC 435), *Enterohacter aerogenes* (MTCC 111) by using disc-agar diffusion method The solutions of ciprofloxacin (antibacterial drug) and clotrimazole (antifungal drug) were used as standard. MICs (minimum inhibitory concentration) of the compounds against test organisms were determined by the broth micro dilution method [xv-xx] and DMSO was used as negative control. All these tests were performed three times under identical conditions and average values were recorded. Activity was determined by measuring the diameter of zone showing complete inhibition and has been expressed in mm.

Synthesis of the Ligands

All the ligands were synthesized in two steps. The first step involved preparation of substituted acetophenones and in second step condensation of acetophenones with glycine.

A) 2-hydroxy-5-methylacetophenone glycine (HMAGLY)
B) 2-hydroxy-5-chloro-4-methylacetophenone glycine (HCMAGLY)
C) 2-hydroxy-5-chloroacetophenone glycine (HCAGLY)

Step I: Preparation of the acetophenones

The acetophenones were prepared by the acetylation of respective substituted phenols following by Fries migration as described below.

a) Preparation of 2-Hydroxy -5 methylacetophenone (HMA).

The acetophenone was prepared by known method (2) it involved the following steps.

i) Preparation of p-Cresyl acetate

p-Cresol(25ml), acetic anhydride (30 ml) and fused sodium acetate (2g) were suspended in 100ml round bottom flask. The mixture was refluxed for 1 h. The content was poured in 500ml water stirring vigorously. The acetic layer was separated and purified by distillation, yield 27.5ml, b.p.208-212^{0}C

ii) Fries' migration of p-cresylacetate

p-Cresylacetate (25ml) and anhydrous aluminum chloride (60g) were taken in Kjeldahl's flask. It was heated in an oil -bath at 115-120^{0}C for 1.25 h After cooling the product was decomposed with dilute hydrochloric acid and ice crashed in an ml breaker. The product was recrystallized from % ethanol, yield 59% m. p. 58^{0}C.

b) Preparation of 2-hydroxy-5-chloro-4-methylacetophenone (HCMA)

The acetophenone was prepared in two steps.

i) Preparation of p-chlorometacresyl acetate

A mixture p-chlorometacresol (25g), acetic acetic anhydride (30ml) and fused sodium acetate (2g) was taken in 250ml round bottom flask. The mixture was refluxed for 1 h. The mixture was cooled to room temperature washed several times with water. Finally, the organic layer was separated and distilled to get the pure acetate, yield 28ml, b. p. 190-192^{0}C.

ii) Fries migration of p-chlorometacresyl acetate

p-Chlorometacresyl acetate (25ml) and anhydrous aluminum chloride (60g) were taken in in Kjeldahl's flask. It was heated in an oil-bath at 115-120^{0}C for 1h. The contents were cooled and decomposed with hydrochloric acid and crushed ice. The product so obtained was recrystallized from 50% ethanol, yield 54%, m. p. 62^{0}C.

c) Preparation of 2-Hydroxy-5-chloroacetophenone (HCA)

The HCA was prepared by known method

i) Preparation of p-Chlorophenyl Acetate

A mixture of p-chlorophenol (25.6g), acetic anhydride (30ml) and fused sodium acetate (2g) was taken in 250ml round bottom flask. It was refluxed for about 1h. The mixture was cooled and poured in to water. It was further purified by distillation, yield 27ml, b. p. 220-222 ^{0}C

ii) Fries' migration of p-chlorophenyl acetate

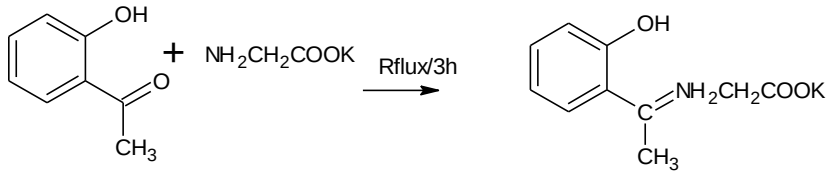
p-Chlorophenyl acetate (20ml) and anhydrous aluminum chloride (48g) were taken in a Kjeldahl's flask. It was heated on an oil -bath at 120-130^{0}C for about 1h. The contents were

cooled and decomposed with dilute hydrochloric acid and crushed ice. The product obtained was recrystallized from 50% ethanol, yield 60%, m. p. 48^0C

Step-II Condensation of acetophenones with amino acid

The amino acid dissolves in a minimal quantity of water. To the hot aqueous solution of amino acid ethanolic solution of KOH were added. The mixture was stirred at room temperature, when the mixture becomes homogeneous, a solution of acetophenone in ethanol was added with constant stirring. The reaction mixture was refluxed on water bath for ~ 3 h and excess of solvent removed by slow evaporation. The separated pale yellow-coloured needles were dried under vacuum over P_2O_5.

A) 2-Hydroxy-5-methylacetophenone glycine (HMAGLY)

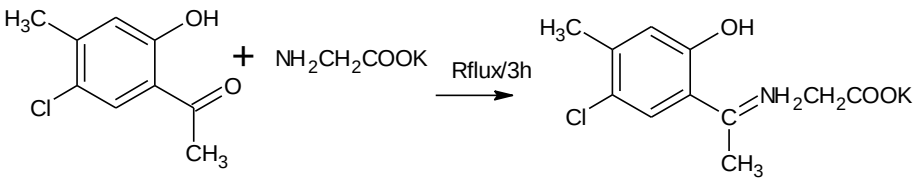


The newly synthesized ligand was characterized with the help of elemental analysis and IR studies. The elemental analysis for carbon, hydrogen and nitrogen is given in table1.The elemental analysis suggests the empirical formula $C_{11}H_{12}NO_3K$ for ligand HMAGLY.

The IR spectrum of ligand HMAGLY shows a medium broad band at 3386 cm^{-1} due to intramolecular hydrogen bonded phenolic υ(OH) and υ(N-H) stretching. These strong band at 1620cm^{-1} is assigned to υ(C=N) stretching frequency [vi]. A band at 1271cm^{-1} is due to υ(C-O) phenolic stretching frequency. The important IR bands of BNPSAP are tabulated in table 2.

^{1}HNMR spectrum of HMAGLY shows expected signals at 5 11.43, (IH, s, -NH); 7.5, 7.1 and 6.9, (3H, m, phenyl); 4.63 (2H, s,-CH2); 2.7, (3H, s, methyl) and 2.4 ppm (3H, s, Ar-methyl) [7,17-18].

B) 2-Hydroxy-5-chloro-4-methylacetophenone glycine (HCMAGLY)

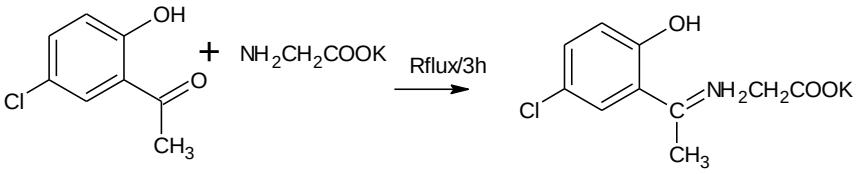


The ligand **HCMAGLY.** was characterized by elemental analysis and IR studies. The elemental analysis of carbon, hydrogen and nitrogen is given in table 1. On the basis of elemental analysis empirical formula $C_{11}H_{11}ClKNO_3$ can be suggested to HCMAGLY**.**

The IR spectrum of ligand shows a medium broad bond at 3401cm$^{-1,}$ which can be assigned to hydrogen bonded phenolic υ(OH) and ν(NH) stretching. The strong band at 1634cm^{-1} is assigned to υ(C=N) group, A strong and sharp band at 1298cm^{-1} is due to υ(C-O) stretching frequency. The assignments of important IR bands are tabulated in table 2.

^{1}HNMR spectrum of HCMAGLY shows expected signals at 6 11.85, (IH, s, -NH); 7.8, 7.1, (2H, s, phenyl); 4.77 (2H, s, -CH2); 2.8, (3H, s, methyl) and 2.5 ppm (3H, s, Ar-methyl).

C) 2-Hydroxy-5-chloroacetophenone glycine (HCAGLY)



The newly prepared ligand HCAGLY was characterized by elemental analysis and IR data. The elemental analysis for carbon, hydrogen and nitrogen is given table 1. From the percentage composition of HCAGLY the empirical formula, which comes out to as $C_{10}H_9ClNO_3K$

The IR spectrum of ligand HCAGLY shows broad band at 3385cm^{-1} due to intramolecular hydrogen bonded phenolic υ(OH) and ν(NH) stretching. A strong band at 1630cm^{-1} indicates the presence of υ(C=N) group and sharp band 1255cm^{-1} may be due to υ(C-O) stretching vibration The assignments of important IR bands are tabulated in table 2.

^{1}H NMR spectrum of HCAGLY exhibits Signals at 6 11.8, (IH, s, -NH); 7.82, 7.7 and 7.67, (3H, m, phenyl); 4.82 (2H, s,-CH2); and 2.85 ppm (3H, s, methyl).

RESULTS AND DISCUSION:

Antimicrobial Activity

The antibacterial activity was evaluated by the single disc method [vii-ix]. The ligands were dissolved in dimethyl sulfoxide at concentration of 10.0mg/ml. The 10mm diameter Whatman no.1 paper disc was soaked in different solutions of the compounds, dried and then placed on the lawn of cultures on nutrient agar plates. The plates were incubated for 24h at 37^{0}C and inhibition zone round each disc was measured. The results were interpreted according to Cappuceino and Sherman method [x]. The ligands were found bactericidal against S. epidermidis, S. flexneri, S. typhi, and S. typhimuriu and bacteriostatic against other strains. The results were

recorded for each tested compound as the average diameter of inhibition zones of bacterial growth surrounding the well in mm. The obtained results are presented in Table 3. and shown in Fig. 1. It is clear that, all of the ligands are more potent bactericides. This difference in the activity probably may be attributed to the fact that the cell wall of Gram +ve bacteria have more antigenic properties as the outer lipid membrane is of polysaccharides, however, their activities were found to be less than the standard ciprofloxacin (antibacterial drug) and clotrimazole (antifungal drug).

CONCLUSION:

In the present study ligands were synthesized and characterized by elemental analysis, melting point, ^{1}NMR, IR, UV-Vis-spectra. The insolubility of the ligands in the organic solvents and the high decomposition temperature indicate their polymeric nature. The antibacterial activity of all the compounds was tested against bacterial pathogens, *E. coli, S. aureus, P. aeruginosa and K pneumoniae*.in fig.no.1. It has been found that synthesized ligands active significant antimicrobial activity.

ACKNOWLEDGEMENTS:

The Authors are also thankful to Sophisticated Analytical Instrument facility, Punjab University, Chandigarh for providing elemental analysis, IR and ^{1}H NMR and ^{13}C NMR facility. Sant Gadge Baba Amravati University, Amravati (Maharashtra) for providing laboratory facilities is also gratefully acknowledged.

REFRENCES:

i. Shivakumar K.; Reddy P.; Vithal R.P.; and Halli M.B.; Synthesis and spectroscopic characterization of metal complexes with naphthofuran-2-carbohydrazide Schiff's base; J. Coord. Chem.; 2007, **60**(3),243-256.

ii. Garzon R.L.; Mascaro's, P.A.; Salido M.L.; Solution study and 2-D layered structures of zinc (II) and cadmium (II) complexes with *N*-2-(6-amino-3,4-dihydro-3-methyl-5-nitroso-4-oxopyrimidinyl)-L-methionine as ligand; Inorganic Chem. Acta.; 2000, **308**,59-64.

iii. Yao K.M.; Zhou W.; Lu G.; Shen L.F.; Huaxue Xuebao; Synthesis, mechanism and NMR spectra of lanthanide complexes with an unsymmetrical Schiff base; Acta Chimica Sinica, ;2000, **58**(10),1275-1279.

iv. Colacio E.; Ghazi M.; Kivekas R.; Moreno; Helical-Chain Copper (II) Complexes and a Cyclic Tetranuclear Copper (II) Complex with Single Syn-Anti Carboxylate Bridges and Ferromagnetic Exchange Interactions; J.M. Inorg. Chem.; 2000, **39**, 2882-2890.

v. Mandlik P. R.; Aswar A. S.; Schiff Base Metal Complexes of Chromium (III), Manganese (III), Iron (III), Oxovanadium (IV), Zirconium (IV) and Dioxouranium (VI); Polish J. Chem.; 2003,**77**,129-135.

vi. Reddy P.R.; Radhika M.; Manjula P.; Synthesis and characterization of mixed ligand complexes of Zn (II) and Co (II) with amino acids: relevance zinc bonding sites in zinc fingers; J. Chem. Sci.;2005, **117(3)**, 239-246.

vii. Aswar A.S.; Mohod R. B.; and Bhadange S.G.; Synthesis and characterization of manganese (III), iron (III), zirconium (IV), thorium (IV), oxovanadium (IV) and dioxouranium (VI) chelates of a polymeric Schiff base.; Indian J. Chem. A; 2001, **40**(10), 1110-1113.

viii. Ilknur Babahan S. E.; Ozturk E.P.; Coban; Spectroscopic and biological studies of new mononuclear metal complexes of a bidentate NN and NO hydrazone–oxime ligand derived from egonol.; Spectrochimica Acta Part A: Molecular and Biomolecular Spectroscopy; 2015, **141**, 300-306.

ix. Beaur A.W.; Kirby W.M.; Sherris J.C.; Turek M.; Antibiotic Susceptibility Testing by a Standardized Single Disk Method.; American Journal of Clinical Pathology.; 1966, **45**(4) 493-496.

x. Cappucino J.C.; Sherman N.; Microbiology: A Laboratory Manual, Addison, Wesley Publishing Co. ;1983.

Received on 15 June 2025

Table 1. Analytical and Physical data of the Ligands

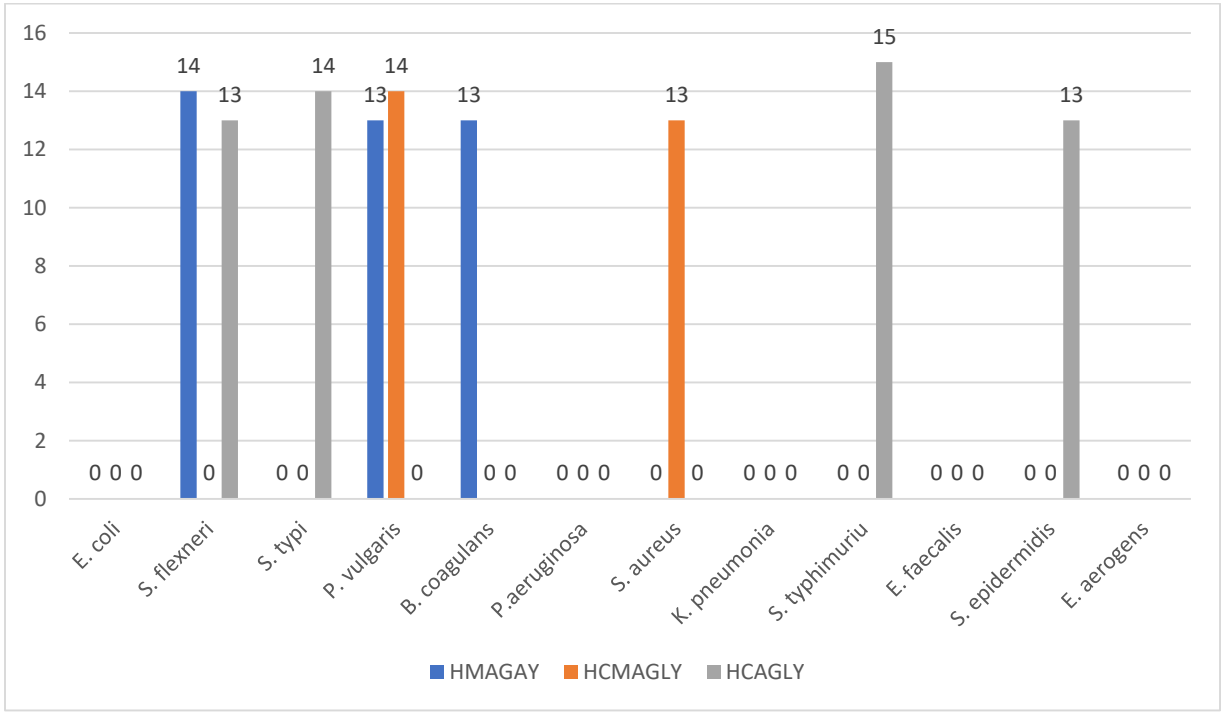
Sr. No.	Ligands	Chemical formula	Molecular Weight	Colour and Nature		Elemental Analysis (%)		
					C Found (calc)	H Found (calc)	N Found (calc)	Cl Found (calc)
1	HMAGAY	$C_{11}H_{12}NO_3K$	245.3	Yellow Crystaline	53.52 (53.86)	5.02 (4.93)	5.65 (5.71)	---
2	HCMAGLY	$C_{11}H_{11}ClKNO_3$	279.3	Yellow Crystaline	46.93 (47.23)	4.18 (3.96)	5.15 (5.27)	12.58 (12.67)
3	HCAGLY	$C_{10}H_9ClNO_3K$	265.7	Dark Yellow Crystaline	48.52 (49.06)	3.35 (3.41)	4.33 (4.77)	13.29 (13.34)

Table 2. The Infrared Spectral data ($cm^{-1)}$ of the ligands

Sr. No.	Ligand	υ(N-H) υ(O-H) cm^{-1}	υ(COO) Assym cm^{-1}	υ(C=O) υ(C=C) cm^{-1}	υ(COO) Symm cm^{-1}	υ(C-O) Phenolic cm^{-1}
1	HMAGAY	3386	1688	1621	1422	1271
2	HCMAGLY	3401	1675	1619	1426	1298
3	HCAGLY	3385	1689	1620	1420	1255

Table 3. Antibacterial activity of Ligands (diameter of inhibition zone in mm)

Ligand	E. coli	S flexneri	S. typi	P. vulgaris	B. coagulans	P. aeruginosa	S. aureus	K. pneumonia	S. typhimuriu	E. faecalis	S. epidermidis	E. aerogens
HMAGAY	R	14	R	13	13	R	R	R	R	R	R	R
HCMAGLY	R	R	R	14	R	R	13	R	R	R	R	R
HCAGLY	R	13	14	R	R	R	R	R	15	R	13	R



Fig,1 Antibacterial activity of Ligands