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SYNTHESIS AND SPECTROSCOPIC CHARACTERIZATION OF TRIDENTATE LIGANDS AND THEIR ANTIMICROBIAL BIOLOGICAL STUDIES

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ABSTRACT:
Schiff base ligand synthesized by condensation of pyrazine-2-carbohydrazide with 1-(5-chloro-2-hydroxyphenyl) ethan-1-one,1-(2-hydroxy-phenyl) ethan-1-one and 1-(2-hydroxy-5-methylphenyl) ethan-1-one were characterized by elemental analyses, magnetic susceptibility measurements, IR, reflectance spectra, thermal analysis, powder X- ray diffraction and SEM analysis. The Schiff base ligands were screened *in vitro* for their biological activity against *E. coli* MTCC 443, *P. aeruginosa* MTCC 424, *S. aureus* MTCC 96, *B. subtilis* MTCC 8979, *E. faecalis* MTCC 439and*S. pyogenes* MTCC 442 and fungal strains, *C. albicans* MTCC 227, *A. niger* MTCC 282 and *A. clavatus* MTCC 1323 and all the ligands. Showed better biological efficacy.

KEYWORDS: Pyrazine-2-carbohydrazone, Biological Activity.

INTRODUCTION:

Hydrazones are characterized by the presence of the triatomic grouping –C=N–N– and are found as interesting ligands in coordination chemistry due to their strong chelating ability through the electron delocalization, which attached with extended conjugation, structure diversity and wide range of possible applications [i-iii.] Hydrazones and hydrazides have also gained considerable interest in recent years owing to their wide variety of biological and pharmacological properties as well [iv-v]. Hydrazone Schiff bases continue to attract attention of several investigators due to their diverse biological applications like antimicrobial [vi] antifungal anticancer [viii] and herbicidal [ix] and so forth. The Schiff bases of hydrazones including heterocyclic moieties involving nitrogen, oxygen and sulphur as coordinating functionalities have been studied extensively in order to establish a relationship between the chemical structure and biological activity [x-xiii].

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.

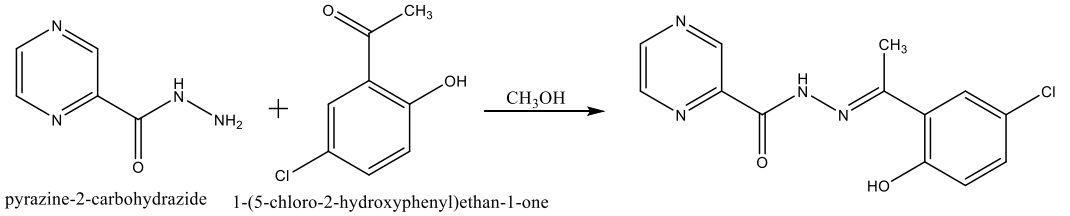
Antimicrobial Activity

The Schiff base ligands (H_2L) were screened for their anti-bacterial and anti-fungal activity against *E. coli* MTCC 443, *P. aeruginosa* MTCC 424, *S. aureus* MTCC 96, *B. subtilis* MTCC 8979, *E. faecalis* MTCC 439and*S. pyogenes* MTCC 442 and fungal strains, *C. albicans* MTCC 227, *A. niger* MTCC 282 and *A. clavatus* MTCC 1323 by using disc-agar diffusion method. [xiv]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 Schiff base ligands

I. **(*N*'-(1-(5-chloro-2-hydroxyphenyl) ethylidene)pyrazine-2-carbohydrazide)**

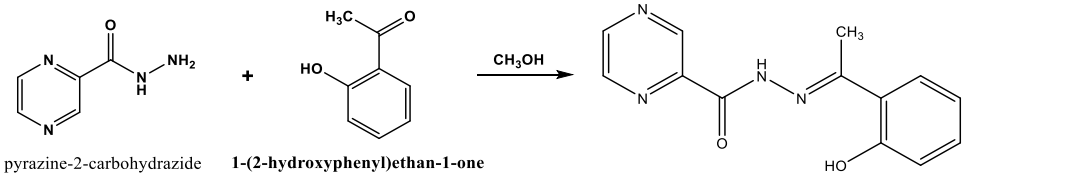
The ligand (*N*'-(1-(5-chloro-2-hydroxyphenyl)ethylidene)pyrazine-2-carbohydrazide) was synthesized by making solution of pyrazine-2-carbohydrazide and 1-(5-chloro-2hydroxyphenyl)ethan-1-one in methanol (30 mL), refluxed in the presence a catalytic amount of glacial acetic acid (1-2 drops) for ca. 4 h on a water bath. After completion of the reaction, the methanol was distilled off using rotary evaporator to nearly half of its volume and then cooled to room temperature which resulted a lemon-yellow solid. This was filtered, washed with hot ethanol and crystallized from DMF. The purity of compound was checked by TLC. Yield 78%, m. p. 248 ^{o}C.



Scheme I. Synthesis of the ligand L^1H

II. ***N*'-(1-(2-hydroxyphenyl) ethylidene) pyrazine-2-carbohydrazide (H_2L)**

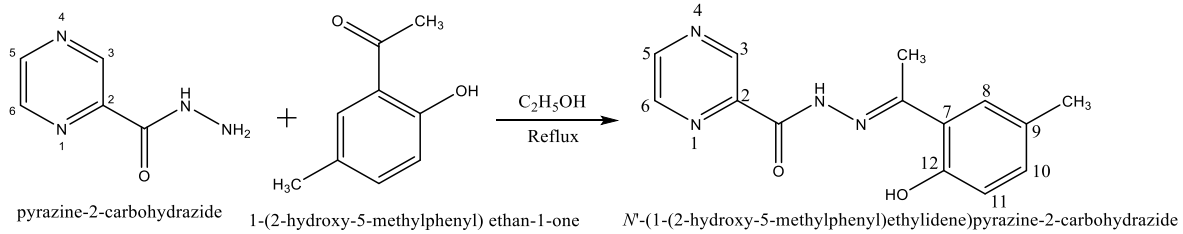
The ligand *N*'-(1-(2-hydroxyphenyl) ethylidene) pyrazine-2-carbohydrazide (H_2L) were synthesis of hydrazone Schiff base was carried out by the condensation of Pyrazine-2-carbohydrazide with 1-(2-hydroxy-phenyl) ethan-1-one in ethanol. The chemical structure of ligand was confirmed by various spectral characterizations (IR, ^{1}H and ^{13}C NMR, and mass spectra). The ligand has been synthesized under double stage synthesis and its synthetic scheme has been shown in scheme II.



Scheme II. Synthesis of the ligand L^2H

III. **1-(2-hydroxy-5-methylphenyl) ethan-1-one pyrazine-2-carbohydrazone (H_2L)**

The synthesis of hydrazone Schiff base 1-(2-hydroxy-5-methylphenyl) ethan-1-one pyrazine-2-carbohydrazone was carried out by the condensation of Pyrazine-2-carbohydrazide with 1-(2-hydroxy-5-methylphenyl) ethan-1-one in ethanol. The resulting reaction mixture was magnetically stirred and refluxed on water bath for 4h and allowed to cool. The progress of reaction was monitored by TLC using silica gel G. The excess of ethanol was evaporated in rotary evaporator to nearly half of its volume and cooled to room temperature which resulted in the formation of a yellow solid product. The yellow-coloured product obtained was filtered, washed with methanol and recrystallized with DMF. The yield and melting point was found to be 78%, and m.p. 235 ^{0}C.



Scheme III. Synthesis of the ligand L^3H.

RESULTS AND DISCUSION:

IR, ^{1}HNMR, 13C NMR, Mass Spectra and XRD

The Schiff base ligand (*N*'-(1-(5-chloro-2-hydroxyphenyl)ethylidene)pyrazine-2-carbohydrazide) L^1H

IR (KBr disc/ cm^{-1}): 3346 (OH), 3182 (NH), 1691 m (C=O), 1630 m (C=N), 1301 (C–O).

^{1}H NMR (DMSO-d_6, 400 MHz): δ 13.11 (s, 1H, OH), 11.60 (s, 1H, NH), 9.28 (s, 1H, J = 1.4 Hz, C3-H), 8.95 (d, 1H, J = 2.4 Hz, C6-H), 8.83 (dd, 1H, J = 1.5, 2.4 Hz, C5-H), 7.67 (d, J = 2.6 Hz. 1H, C6'-H), 7.36 (dd, 1H, J = 2.6, 8.8 Hz, C4'-H), 6.95 (d, 1H, J = 8.8 Hz, C3'-H), 2.51 (s, 3H, CH_3)

^{13}C NMR (DMSO-d_6, 100 MHz): 160.17 (C=N), 158.97 (C2'), 157.40 (C=O), 148.10 (C6), 144.14 (C3 & C5), 143.57 (C2), 131.15 (C4'), 128.0 (C6'), 122.29 (C5'), 119.13 (C3'), 120.73 (C1'), 14.10 (CH_3)

The Schiff base ligand *N*'-(1-(2-hydroxyphenyl)ethylidene)pyrazine-2-carbohydrazide L^2H Micro analytical data for $C_{13}H_{12}N_4O_2$. Anal. Calc., C, 60.93; H, 4.72; N, 21.86. Found C, 61.12; H, 4.70; N, 21.83%. IR (KBr disc, cm^{-1}), 3346(OH), 3182 (NH), 1691 m(C=O), 1630 m(C=N), 1301 (C–O).

The Schiff base ligand 1-(2-hydroxy-5-methylphenyl) ethan-1-one pyrazine-2-carbohydrazone L^3H

The molecular weight of the ligand was confirmed by mass spectral data. The formation of ligand was confirmed by ^{13}Cand ^{1}H- NMR spectral data.

IR (KBr disc, cm^{-1}): 3338(OH), 3178 (NH), 1688 (C=O), 1626 (C=N), 1298 (C–O)

^{1}H- NMR (DMSO-d_6, 400 MHz): δ12.61 (s,1H, OH), 11.29 (s,1H, NH), 9.32 (s, 1H, C3-H), 8.89 (d, 1H, C5-H), 8.73 (s, 1H, C6-H), 7.36 (s, 1H, C8-H), 7.09 (d, 1H, C10-H), 6.79 (m, 1H, C11-H), 2.50 (s, 3H, CH_3)), 2.27 (s, 3H, CH_3).
^{13}C NMR (DMSO-d_6, 100 MHz): 161.76(C=N), 159.27(C12), 156.55(C=O), 147.75(C6), 143.99(C2), 143.66(C3), 142.89(C5), 132.09(C10), 128.19(C9), 126.97(C8), 118.41(C7), 117.10(C11), 20.20(CH_3), 13.37(CH_3).
Mass spectrum (ESI) = 271.2 [M+1], 272.2 [M+2].

Antimicrobial Activity

All the newly synthesized compounds were screened *invitro* for their antibacterial and antifungal activities against bacterial strains *S. aureus* MTCC 96, *S. pyogenes* MTCC 442, *B. subtilis* MTCC 8979, *E. coli* MTCC 443, *P. aeruginosa* MTCC 424 &*E. faecalis* MTCC 439 and fungal stains *A. niger* MTCC 282, *A. clavatus* MTCC 1323, *C. albicans* MTCC 227. 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 (H_2L) 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 Schiff base ligands were synthesized and characterized by elemental analysis, melting point, ^{1}NMR, IR, UV-Vis-spectra. The ligands were insoluble in the organic solvents. 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 Schiff base show significant antimicrobial activity.

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REFRENCES:

i. Ribeiro N.; Galvao A. M.; Gomes C. S. B; Naphthoyl hydrazones: coordination to metal ions and biological screening, New J. Chem.;2019, **43**, 17801-17818.
ii. Dhanaraj C.J;, Johnson J. ; Joseph J.; Quinoxaline-based Schiff base transition metal complexes, J. Coord. Chemistry; 2013, **66** , 1416-1422.
iii. Dikio C.W.; Ejidike I.P.; Mtunzi F.M.; Klink M.J.; Dikio E.; Hydrazide Schiff bases of acetylacetonate metal complexes: Synthesis, spectroscopic and biological studies, International Journal of Pharmacy and Pharmaceutical Sciences; 2017, 9(12), 257-267.
iv. Kirthan B.R.; Prabhakaraa M.C.; Bhojya Naik H.S; Viswanath R.; Amith Nayak; P.H.; Optoelectronic, photocatalytic and biological studies of mixed ligand Cd (II) complex and its fabricated CdO nanoparticles, Journal of Molecular Structure; 2021, **1244(15),** 130917.
v. Ahmad Desoky M. ; Mohamad M.J.A.; Abualreish Ahmed M. ;Abu-Dief, Antimicrobial and anticancer activities of cobalt (III)-hydrazone complexes: Solubilities and chemical potentials of transfer in different organic co-solvent-water mixtures, Journal of Molecular Liquids;2019, 290(15), 111162.

vi. Altntop M. D.; A.Ozdemir ; Turan-Zitouni ; Synthesis and biological evaluation of some hydrazone derivatives as new anticandidal and anticancer agents, Eur. J. Med. Chem.; 2012, **58**.299-307.

vii. Goff G.L; Ouazzani J.; Natural hydrazine-containing compounds: Biosynthesis, isolation, biological activities and synthesis, Bioorg. Med.Chem,; 2014, **22(23)** ,6529-6544.

viii. A.S.El-Tabl; M. M.; Abd-El Wahed M.A.;Wahba, M.A.Shebl ;Spectroscopic characterization and cytotoxic activity of new metal complexes derived from (1e, n'z, n',n'-bis(2- hydroxybenzylidene)-2-(naphthalen-1-yloxy) acetohydrazonohydrazide, Journal of Advances in Chemistry, ;2015 .**11(9),** 3888-3918.

ix. Sebastian O.; Thapa A.; Schiff base metal complexes of Ni, Pd and Cu, J. Chem. Pharm. Res, ; 2015 , **7(10),** 953-963.

x. El-Halim H. F. ; Mohamed G. G;. Khalil A. M. E; Synthesis, spectral, thermal and biological studies of mixed ligand complexes with newly prepared Schiff base and 1, 10-phenanthroline ligands, J. Mol. Structure; 2017, **1146** (2017), 153-163.

xi. Hossain , M .S. ; Roy P. K. ; Zakaria C. M. ; Selected Schiff base coordination complexes and their microbial application,Int. J. Chemical Studies, ; 2018, **6(1)** ,19-31.

xii. A.S.El-Tabl; M. M.; Abd-El Wahed; Wahba M. A. ; Shebl M. A.; Spectroscopic characterization and cytotoxic activity of new metal complexes derived from (1e, n'z, n'z)-n',n'-bis(2-hydroxybenzylidene)-2-(naphthalen-1-yloxy)acetohydrazonohydrazide, Journal of Advances in Chemistry; 2015. **11(9)** ,3888-3918.

xiii. Bradan G.; Cobeljic B.; Pevec A.; Synthesis, characterization and antimicrobial activity of pentagonal-bipyramidal isothiocyanato Co (II) and Ni (II) complexes with 2,6 diacetylpyridine bis(trimethylammoniumacetohydrazone, J. Coord. Chem; 2016 ,69, 801-811.

xiv. Bhaskar R. S.; Ladole C. A; Salunkhe N. G. ; Aswar A. S.; Synthesis, Characterization and Biological Studies of Some Transition Metal Complexes with Pyrazine Schiff Base Hydrazone Ligand, Jordan Journal of Chemistry, ;2020,**5(2)**, 61- 72.

xv. Gross D.C.; De-Vay J.E.; Production and purification of syringomycin, a phytotoxin produced by *Pseudomonas syringae*, Physiol. Plant Pathol ; 1977, **11,**13-28

xvi. Ahmed M.Abu-Dief, Nashwa M.El-Metwaly ; Seraj OmarAlzahrani ; FatmahAlkhatib , Matokah M.Abualnaja; TarekEl-Dabea; Mahmoud Abd El Aleem Ali AliEl-Remaily; Synthesis and characterization of Fe(III), Pd(II) and Cu(II)-thiazole complexes; DFT, pharmacophore modeling, *in-vitro* assay and DNA binding studies, Journal of Molecular Liquids; 2021, **326**, 115277.

xvii. Ahmed M.Abu-Dief**;** Laila H. Abdel-Rahman**;** Antar A.Abdelhamid; Adel A.Marzouk; Mohamed R.Shehata**;** Mohamed AbdelaalBakheet**;** Omar A.Almaghrabi**;**AymanNafady;Synthesis and characterization of new Cr(III), Fe(III) and Cu(II) complexes incorporating multi-substituted aryl imidazole ligand: Structural, DFT, DNA binding, and biological implications, Spectrochimica Acta Part A: Molecular and Biomolecular Spectroscopy; 2020, **228(5),** 117700.

xviii. Ahmed M. Abu-Dief; Hussein M. El-Sagher, Mohamed R; Shehata; Fabrication, spectroscopic characterization, calf thymus DNA binding investigation, antioxidant and anticancer activities of some antibiotic azomethine Cu (II), Pd (II), Zn (II) and Cr (III) complexes, Applied Organometallic Chemistry; 2019, 33 **(8)**,4943.

xix. Ahmed M.Abu-Dief;Laila H.Abdel-Rahman, Mohamed R.Shehata; Azza A. HassanAbdel-Mawgoud; Novel azomethine Pd (II)- and VO (II)-based metallo-pharmaceuticals as anticancer, antimicrobial, and antioxidant agents: Design, structural inspection, DFT investigation, and DNA interaction, Physical Organic Chemistry; 2019, 32 (12), 17-34.

xx. Maurya R.C.; Singh H.; Singh T.; Metal chelates of bioinorganic and catalytic relevance: Synthesis, magnetic and spectral studies of some mononuclear and binuclear oxovanadium (IV) and dioxotungsten (VI) complexes involving Schiff bases derived from 4-butyryl-3- methyl-1-phenyl-2-pyrazolin-5-one and certain aromatic amines. *Indian J. Chem. A.* 2001**,40**,1053-1063.

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Table 1. Analytical and physical data of the ligands.

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Compounds	Chemical Formula	Molecular Weight	Yields (%)		Elemental Analysis (%)		
				C Found (calc)	H Found (calc)	N Found (calc)	Cl Found (calc)
L^1H	$C_{13}H_{13}ClN_4O_2$	292.70	78	52.98 (53.34)	4.14 (4.48)	18.98 (19.14)	12.00 (12.11)
L^2H	$C_{13}H_{12}N_4O_2$	256.25	80	61.12 (60.93)	4.70 (4.72	21.83 (21.86)	--
L^3H	$C_{14}H_{14}N_4O_2$	270.29	79	61.86 (62.21)	4.98 (5.22)	20.24 (20.73)	--

Table 2. Infrared spectral bands (cm^{-1}) of the ligands.

Compounds	ν(OH-N)	ν(NH)	ν(C=O)	ν(C=N)	ν(C–O) phenolic	ν(N–N)
L^1H	3346	3182	1691	1630	1301	977
L^2H	3342	3180	1672	1626	1294	968
L^3H	3338	3178	1688	1626	1298	970

Table 3. Antimicrobial activity of the ligands.

Compounds	*S. aureus* (mm)	*B. subtilis* (mm)	*E. coli* (mm)	*E. faecalis* (mm)	*A. niger* (mm)	*C. albicans* (mm)
L^1H	12	11	11	12	12	10
L^2H	12	12	13	11	12	08
L^3H	12	14	12	13	12	12
ciprofloxacin	25	24	20	24	--	--
Ketoconazole	--	--	--	--	24	25

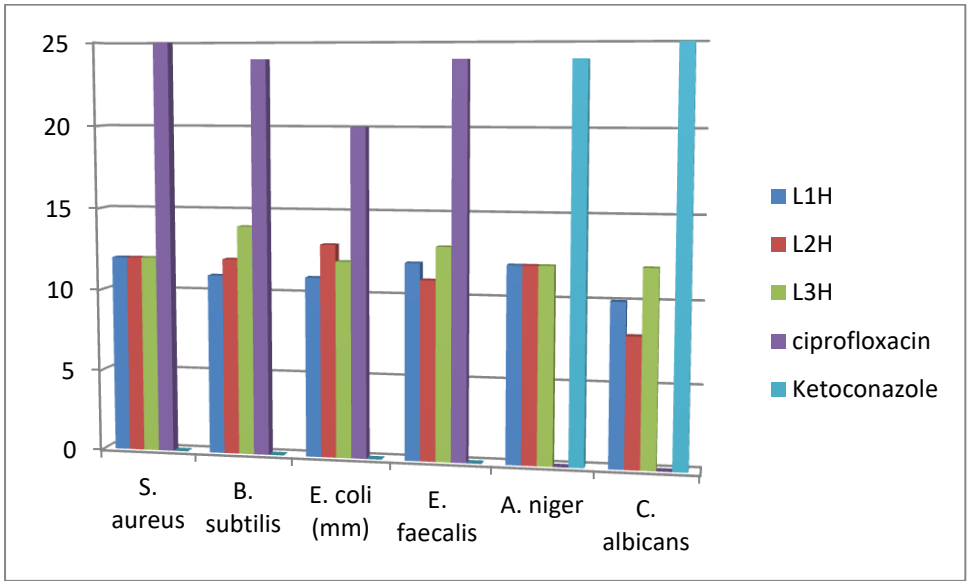

Fig.1 Antibacterial and Antifungal activity of the ligands.