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Synthesis of Schiff base Ligand through Sustainable methods and study of antibacterial properties

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Abstract

Ligands are fascinating class of ions or molecule that binds to a central metal ion to form coordination compounds. Ligand ability to donate lone pair of electrons or capacity to act as"Lewis Bases" has created tremendous wave in pharmaceutical industry. They are multifaceted class of compound formed by condensation of aldehyde or ketone with a primary amine under preliminary condition. The ligands are efficient enough to act as antibacterial, antiviral, anti-inflammatory, antiproliferative antifungal, anticancer properties. In vitro antimicrobial effect of Schiff bases encourages the researchers for the development of new anti-biofoulants products. Ligand preparation is accomplished under mild conditions with reasonably good to high yield. Organometallic complexes offer potential for the development of new materials with a wide range of applications. Due to electronic transitions which exists between Ligand and metal complexes it provides a platform for drug design that is exploited in several areas. The synthesized Alloxan 2,4 dinitrophenyl hydrazone using IR, NMR and screened for their antibacterial . The ligands are tested for their antibacterial activities on gram-positive and gram-negative bacteria.

Key words: Schiff Base, Alloxan 2,4 dinitrophenyl hydrazone IR, NMR, Antibacterial

1. Introduction:

Schiff base named after the scientist Hugo Schiff base are milder, efficient, less hazardous leading to synthesis of variety of Schiff's base. They are synonymous to azomethine (RCH=NR') formed by nucleophilic condensation of carbonyl compounds with primary amines in presence of

acid or base. Hydrogen bonding ability, proton transfer equilibria offer wide variety of biological activity. The magnitude of antibacterial resistance requires discovery of new drug which can be synthesized in an environmental friendly manner leading to a sustainable development [1-5].

Sustainable development meets the needs of the present without compromising the ability of future generations to meet their own needs. Chemists think of sustainable development in terms of preserving environment for the future. Disposal of hazardous chemical waste can be prevented by neutralizing chemicals to appropriate pH or green method of synthesis can be designed leading to sustainable development. In present work we present the synthesis, characterization, biological activity of Schiff's base Ligand synthesized. Acid catalysed condensation reaction of carbonyl compounds with amines is carried out using citric acid (lemon) leading to green technology.

Slightly modified structures of parent Schiff bases leads to enhance activity and reduce side effects relative to the parent molecule. The momentum in utilizing bioactive starting material those have vicinal carbonyl group on either side can provide bio- active sites. Alloxan- 2,4,5,6 pyrimidine tetrone is a heterocyclic compound with high biological and physiological effect on living organisms. Alloxan is reported as an agent which selectively destroys pancreatic beta cells of mice which result in inducing permanent diabetes. The compound that selectively damage β cells constitute diabetogenic drugs. Evidence says that pancreatic β cell damage induced by Alloxan is through generation of cytotoxic oxygen free radicals [6-10].

Knowing the extensive application of Schiff base Alloxan 2, 4, dinitro phenyl hydrazone ligands was synthesized in an eco friendly manner. Condenstion of the Alloxan (keto) with Alloxan 2, 4 dinitrophenyl hydrazine was carried out using citric acid in place of acetic acid. The reaction was carried out in microwave assisted reaction. Its characterization was done using UV, CHN,IR , NMR[11-15].In our present study we aim at studying the biological efficacy of the multifaceted ligand Alloxan-2,4 dinitrophenyl hydrazone derivatives and its Antimicrobial property is studied.

2. Experimental Section:

2. Synthesis of Ligand: The ligand was synthesized using the reagent and solvents purchased from Sigma Aldrich, (Merck), Loba-chemie etc. and these were used without any further

purification. The melting point of the compounds was determined by an open capillary thermal melting point apparatus. FT-IR spectra were recorded as KBr pellets on a Bruker FT-IR spectrophotometer. 1HNMR spectra were recorded in DMSO-d6 on JEOL Delta-550 spectrometer (400 MHz).

2.1 The ligand Alloxan 2,4,dinitrophenyl hydrazine was synthesized using 0.01M solution of Alloxan in methanol refluxed with 0.01M solution of 2,4 dinitrophenyl hydrazine in ethanol for three hours at 70-80℃.Cooling the reaction mixture yielded affordable precipitate. Filtered the yellow coloured precipitate which was recrystallised using ethanol where orange crystals, were obtained.

3. Antimicrobial sensitivity test

3.1 Anti Bacterial Activity:

Well diffusion method to determine the Minimum Inhibition Concentration (MIC)

The antimicrobial assay was determined byWell diffusion methods against all the compounds on Luria Bertani(LB) agar media (Himedia Ltd). The samples were assessed for their MIC property against organisms *Bacillus cereus, E-coli and Pseudomonas aeruginosa.* Zone of the inhibition was measured in mm after the incubation period.

Sample preparation

10 mg each of the samples were dissolved in 1mL of Dimethyl sulfoxide (DMSO). Different aliquots of the sample containing 100µg, 200µg, 300µg, and 400µg was prepared by pipetting 10µL, 20µL, 30µL and 40µL and the final volume was made upto 50µL by adding DMSO.

Media preparation

Luria Bertani (LB) agar media (tryptone 10g, sodium chloride 10g, yeast extract 6g, agar 15g, distil water 1000mL) was prepared and autoclaved at 121°C for 15 mins.

3.2 Plate preparation

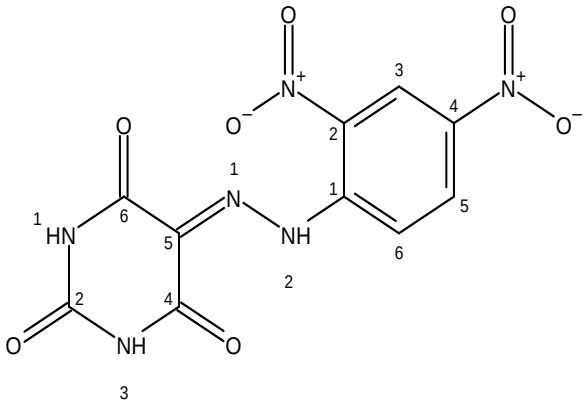
Approximately 25mL of the media was poured into the sterilized petriplates and allowed to solidify. Later 24hrs cultured 100µL inoculum of *Bacillus cereus, E-coli, Pseudomonas aeruginosa* were added into the respective plates and spread throughout the plate using spreader.

Five wells were punctured using well borer and the samples containing 100µg, 200µg, 300µg, and 400µg were loaded into the respective wells and 50µL of DMSO loaded in the center well as control blank and incubated at 37°C for 24hrs.

4. Results and Discussion:

4.1 Elemental analysis

The ligands are variously coloured crystalline powders are obtained, air stable insoluble in common organic solvents but soluble in DMSO. The results of elemental analysis along with molecular formula and melting points are determined. The melting point of the L_1 was found to 250℃ respectively. Analytical data is represented in Table 1.



5-(2-(2,4-dinitrophenyl)hydrazineylidene)pyrimidine-2,4,6 (1H,3H,5H)-trione

Table1. Analytical data of L_1

Compound	Colour	M.P (°C)	Found (Calc)%				Λ_M^*
			C	H	N	S	
Alloxan-2,4 DNPH	Red	250	37.84 (37.26)	1.22 (1.86)	25.79 (26.08)	-	13.6

4.2 FTIR spectral studies:

The IR spectral bands of ligand long with assignments are presented in Table 2. The analysis of IR spectrum of pure ligand (L_1) Figure 1 reveals the Schiff base formation with a band appearing in the region 1620-1667cm^{-1}(Monica.B et al.., 2003). The presence of N-H asymmetric stretching vibration in alloxan, showing absorption bands appearing at ~3500, 3491,

and 3500 cm $^{-1}$ appeared in ligand and complexes. The absorption band at 1528 cm^{-1}-1500 cm^{-1} to symmetric stretching vibration of −N−O due to nitro group present in benzene ring. Ring υ (C=O) due to Alloxan 1715 to 1740cm^{-1} (Kovalchukova.O.V et al.., 1981; Leon Palomino.M.I et al.., 1981).The band at 1390 due to C-NH is lowered to 1377,1370cm^{-1} suggesting coordination through nitrogen of 2,4 DNPH.

Table2 FTIR OF L_1

L_1 (cm^{-1})	N-H	C=N	Aromatic C-H ring	C-N Aromatic	N-O	C-NH
ADNPH	**3500**	**1620**	**3010**	**1212**	**1500**	**1390**

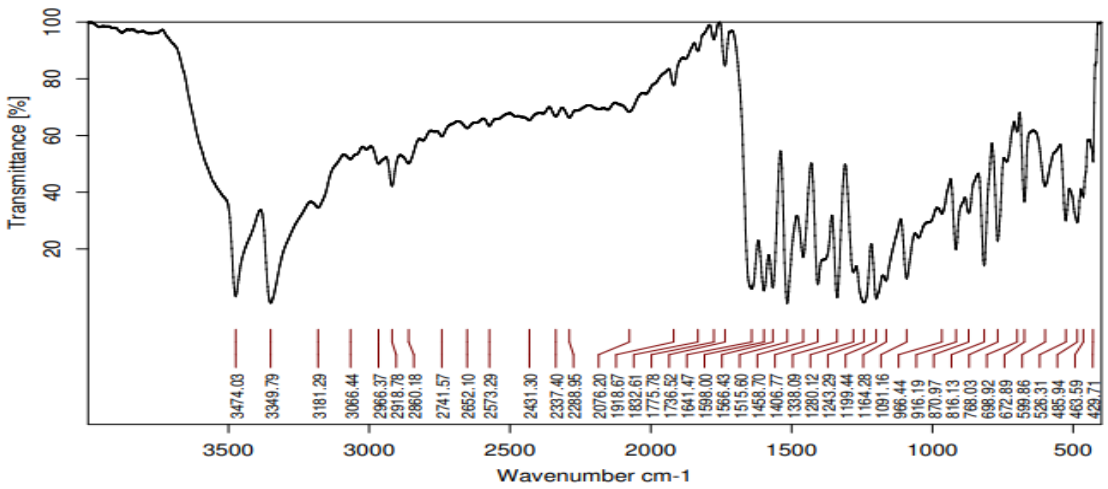


Figure1 :FTIR of L_1

4.3^{1}H NMR spectra: ^{1}H NMR (DMSO-d6) spectrum of the ligands displays the following signals ^{1}H NMR (DMSO-d6) spectrum of the ligands displays Table 3. There is a downfield shift at 11.87- 11.62 ppm (s) (L_3) due to imino proton bound to pyramidine (alloxan) ring integrate as single proton (Offiong et al.., 1995). Furthermore signal at 15.25 ppm (L_1) indicates a down field

shift due to NH (hydrazinic proton) of DNPH. Peaks at 8.94 (d) 8.67(d),8.25(d) correspond to CH proton of phenyl environment proton.

Table3: ^{1}H NMR for the L_1

S.No	Ligand Formula	Alloxan ring N-H proton	Ring CH proton	NH proton of hydrazine
1.	$C_{10}H_6N_6O_7$	11.87,11.62	8.94 (d)8.6(d) 8.25(d)	15.25

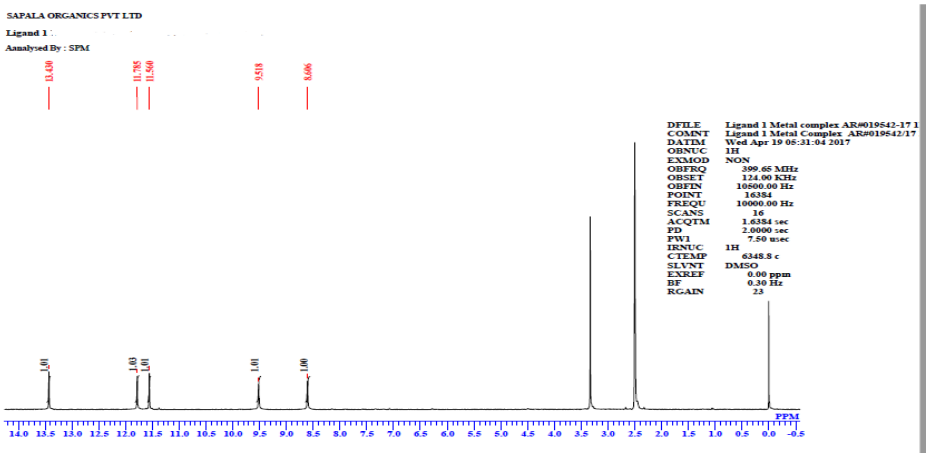


Figure2: NMR spectra L_1

5.1 Antibacterial Activity

Antimicrobial activity was tested against a standard Tetracycline is an antibiotic fights against infections caused by bacteria that affects skin,,intestine, urinary tracts etc. Figure (3) indicates MIC activity of the ligand. It was found to be more effective against bacteria E.coli responsible for urinary tract infections. Since the ligand show zone of inhibition in (cm) can be treated as an effective bacteriostatic agent against E.coli. The primary concept underlying the quantification of the compound antimicrobial assay is to identify and analyze the growth or inhibition of unaltered bacteria, while confirming the absence of any adverse effects. In theory, the growth of bacteria can be inhibited (bacteriostatic) if ionizing disinfectant molecules are absorbed or displaced by

electrical charges during the initial contact and absorption phase. The antibacterial effects of the ligand can be attributed to its ability to penetrate the lipophilic membrane of the bacterium being studied.

Table 3: Antibacterial Activity L_1

	Micro organism zone of inhibition (in cm)		
Concentration	Bacillus cereus	Escherichia coli	Pseudomonas aeruginosa
100 µg	-	-	-
200 µg	-	1.0	-
300 µg	-	1.2	-
400 µg	0.9	1.3	1.2

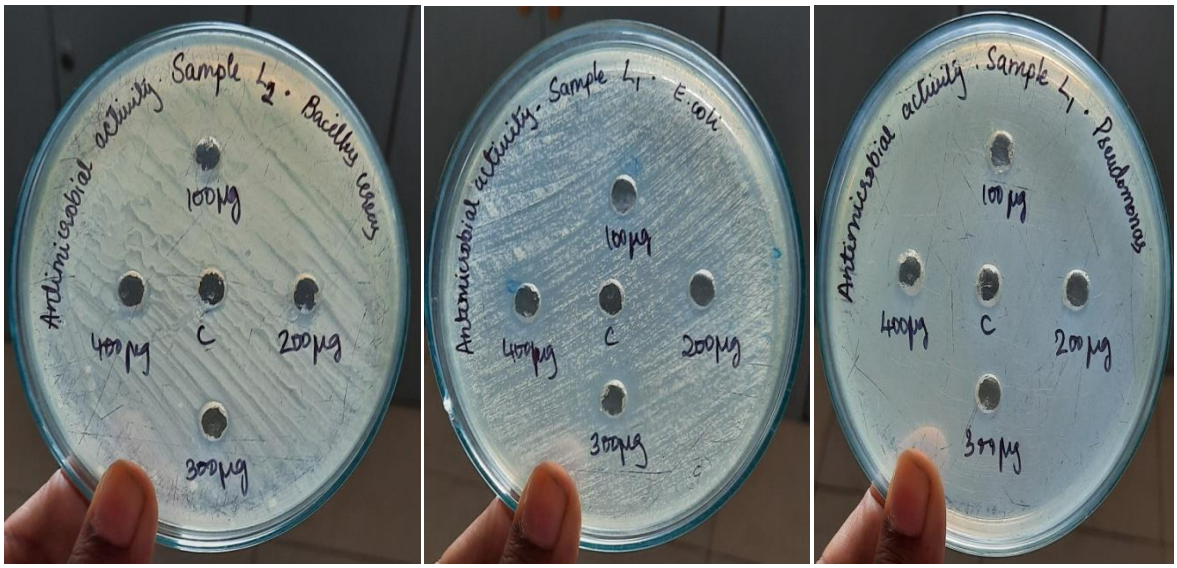


Figure3: Zone of Inhibition of L_1

6. Conclusion:

Green method of synthesis of the ligand was found to be effective leading to a sustainable development. Futuristic work of the ligand involves synthesis of complexes and its characterization. Cell line studies of the synthesised ligand and complex can be studied for their antitumour properties.

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8. References:

1. Padhye, S., & Kauffman, G. B. (1985). Transition metal complexes of semicarbazones and thiosemicarbazones. *Coordination Chemistry Reviews*, *63*, 127-160.

2. Carter, J. W., Mayes, R., Pierce, K. A., Lawson, R., Lisic, E. C., & Edward, C. (2003). Structural determination of a series of ortho-quinone thiosemicarbazone compounds using NMR spectroscopy. *J. Und. Chem. Res*, *2*, 73-77.

3. Lisic, E. C., Nareddy, R. R., Huxford, R., & Lisict, E. C. synthesis of a series of potential antitumor metal-complexing ligands: alloxan thiosemicarbazone, alloxansemicarbazone compounds. *Ariel*, *130*, 248-134.
4. Patil, R., Patil, R., Ahirwar, B., & Ahirwar, D. (2010). Antidiabetic activity of leaves of jatropha curcus l.(euphorbiaceae) in alloxan induced diabetes rats. *Pharmacology online*, *3*, 662-8.

5. Cohen, G., & Cederbaum, A. I. (1980). Microsomal metabolism of hydroxyl radical scavenging agents: relationship to the microsomal oxidation of alcohols. *Archives of Biochemistry and Biophysics*, *199*(2), 438-447.

6. Padhye, S., Afrasiabi, Z., Sinn, E., Fok, J., Mehta, K., & Rath, N. (2005). Antitumor metallothiosemicarbazonates: structure and antitumor activity of palladium complex of phenanthrenequinone thiosemicarbazone. Inorganic Chemistry, 44(5), 1154-1156.

7. Rho, H. W., Lee, J. N., Kim, H. R., Park, B. H., & Park, J. W. (2000). Protective mechanism of glucose against alloxan-induced β-cell damage: Pivotal role of ATP. Experimental & Molecular Medicine, 32(1), 12-17.

8. Kumari, Y. P., Chandra, J. S., Rao, B. S., & Sunandamma, Y. (2012). Synthesis, characterization and antibacterial activity of Alloxanthiosemicarbazone Au (III) complexes. *Journal of Current Pharmaceutical Research*, *10*(1), 28-33.

9. Beraldo, H., & Gambino, D. (2004). The wide pharmacological versatility of semicarbazones, thiosemicarba-zones and their metal complexes. Mini reviews in medicinal chemistry, 4(1), 31-39.

10. Bojarski, J. T., Mokrosz, J. L., Bartoń, H. J., & Paluchowska, M. H. (1985). Recent progress in barbituric acid chemistry. Advances in heterocyclic chemistry, 38, 229-297.

11. Shebaldina, L. S., Kovalchukova, O. V., Strashnova, S. B., Zaitsev, B. E., & Ivanova, T. M. (2004). Synthesis and Physicochemical Properties of d-and f-Metal Complexes with Alloxan. *Russian Journal of Coordination Chemistry*, 30, 38-42.

12. Kannan, S., & Ramesh, R. (2006). Synthesis, characterization, catalytic oxidation and biological activity of ruthenium (III) Schiff base complexes derived from 3-acetyl-6-methyl-2H-pyran-2, 4 (3H)-dione. *Polyhedron*, *25*(16), 3095-3103.

13. He, Y., Wu, B., Yang, J., Robinson, D., Risen, L., Ranken, R., & Swayze, E. E. (2003). 2-Piperidin-4-yl-benzimidazoles with broad spectrum antibacterial activities. *Bioorganic & medicinal chemistry letters*, *13*(19), 3253-3256.

14. Klimešová, V., Kočí, J., Waisser, K., & Kaustová, J. (2002). New benzimidazole derivatives as antimycobacterial agents. *Il Farmaco*, *57*(4), 259-265.

15. Horton, D. A., Bourne, G. T., & Smythe, M. L. (2003). The combinatorial synthesis of bicyclic privileged structures or privileged substructures. *Chemical reviews*, *103*(3), 893-930.