

# Synthesis, Characterization of Silver Nanoparticle Coated With Schiff Base and Their Antibacterial Studies

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**Abstract-** The Schiff base named *N*-(2-hydroxybenzylidene) - 2- amino pyridine [HBAP] formed from condensation of equimolar (1:1) quantity of salicylaldehyde and 2- amino pyridine in alcoholic medium. The prepared Schiff base was characterized by elemental analysis, FTIR, UV-VIS, NMR spectral studies. Synthesis of silver nanoparticles followed by coating of silver nanoparticles with above mentioned Schiff base. The structure of silver nanoparticle anchored with Schiff base is determined by UV-VIS spectra. Antibacterial activity of test compounds were assessed against gram positive bacteria such as *S.aureus*, *M.luteus*, *B.licheniformis* and gram negative bacteria such as *E.coli* by paper disc method and results were compared with the antibiotic, ofloxacin. The antibacterial study revealed that silver nanoparticle coated with Schiff base showed excellent activity against gram positive bacteria: *S.aureus*, *M.luteus*, *B.licheniformis* and gram negative bacteria: *E.coli*.

**Keywords-** Schiff base, silver nanoparticles, antibacterial properties, ofloxacin.

## I. INTRODUCTION

New areas of researches, which mainly focus on the synthesis specific, highly functional metal drug complexes, have drawn considerable attention. Schiff base known as azomethine derivative and it is an important class of ligand that coordinated with metal ion via nitrogen of azomethine. Schiff base was versatile ligand and shows antibacterial, antifungal, anticancer and diuretic activities [1]. For showing these properties Schiff base is widely used in food industry, dye industry, analytical chemistry, catalysis, fungicidal, agrochemical and biological activities [2]. Nanotechnology is the study of materials smaller than 100 nanometres (nm) [3]. Silver nanoparticles have been used since ancient times for jewellery, utensils, monetary currency, dental alloy, photography, explosives, etc. Until the introduction of antibiotics, it was also used for its antiseptic activity, specifically in the management of open wounds and burns. Silver has also been used in filters to purify drinking water and

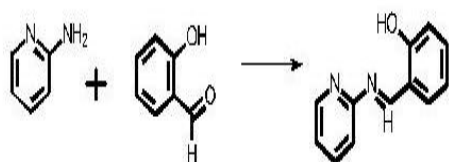
clean swimming pool water due to its antimicrobial properties. The antimicrobial drugs occupy a unique niche in the history of medicine. Considering the increased incidences of severe opportunistic bacterial infections in immunological deficient patients together with the development of resistance among pathogenic gram positive and gram negative bacteria, there is a great need in finding new compounds that may be effective against antibiotic resistant bacteria. During the entire preceding history of medicine fewer of drugs had known focus of action and then fewer had been submitted to synthetic investigations. In the present work, The Schiff base named *N*-(2 hydroxybenzylidene) - 2- amino pyridine [HBAP] formed from condensation of equimolar (1:1) quantity of salicylaldehyde and 2- amino pyridine in alcoholic medium. The prepared Schiff base was characterized by elemental analysis, FTIR, UV-VIS, NMR spectral studies. Silver Nanoparticles were synthesized by chemical reduction method. Silver nitrate used as a metal precursor and sodium borohydride as a reducing agent. Synthesis of silver nanoparticles followed by complexation of silver nanoparticles with above mentioned Schiff base. The structure of silver nanocomplex of Schiff base is determined by UV-VIS spectra and further screened for antibacterial activity against gram positive bacteria such as *S.aureus*, *M.luteus*, *B.licheniformis* and gram negative bacteria such as *E.coli* by using paper disc method. In an attempt to identify a potent and safer antimicrobial agent, we focused our efforts towards the synthesis of novel silver nanoparticle coated Schiff base.

## II. MATERIAL AND METHODS

**MATERIAL:** All chemicals and solvent used were of analytical grade. UV-VIS spectra were obtained on digital spectrophotometer in the range 300-900nm in DMF. IR spectra were recorded using KBR disc on a FT-IR spectrophotometer, Shimadzu 8201PC in the range of 4000-400cm<sup>-1</sup>. <sup>1</sup>HNMR spectra were recorded in MeOD at room temperature. Elemental analysis was carried out on Elementar Vario ELIII. Melting point of the Schiff base was determined

by open capillary method using sunsim electric melting point apparatus.

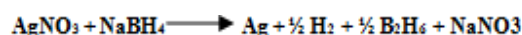
**SYNTHESIS OF SCHIFF BASE:** A solution of salicylaldehyde in ethanol mixed with solution of 2-amino pyridine and adds few drops of formic acid. Mixture is stirred and thereafter refluxed for 7-8 hrs, the precipitate collected by filtration and recrystallized from alcohol.



2-amino pyridine salicylaldehyde HBAP

**SYNTHESIS OF SILVER NANOPARTICLE:** Fixed volume of silver nitrate solution added drop wise to ice chilled sodium borohydride solution. The reaction mixture was stirred vigorously on magnetic stirrer. The entire addition took about three to four minutes, after which the stirring was stopped and the stir bar removed. The colloidal solution of nanoparticle was formed.

**Chemical reduction method:**



**COATING OF SILVER NANOPARTICLE BY SCHIFF BASE:** coating of silver nanoparticle by Schiff base was done by anchoring the ligand as prepared above on the synthesized silver nanoparticles [4]. Schiff base was successively added to the silver nano colloidal solution in and stirred for 10-15 minutes.

**ANTIBACTERIAL STUDIES:** Evaluation of antimicrobial activity of all compounds *in vitro* was carried out by paper disc method against bacteria including *E. coli*, *S. aureus*, *M. luteus*, and *B. lichenformis*. Ofloxacin was additionally tested as positive control.

### III. RESULT AND DISCUSSION

#### CHARACTERIZATION OF SCHIFF BASE:

Schiff base colour, yield melting point, elemental analysis was shown in table-1.

Table-1 Physical properties of Schiff base

Colour	Yellow orange crystal
Yield	57%
Melting point	62-64°C
Anal. Calculated for C <sub>12</sub> H <sub>10</sub> N <sub>2</sub> O	C- 72.75, H-5.02, N-14.14
Found for C <sub>12</sub> H <sub>10</sub> N <sub>2</sub> O	C- 72.30, H-5.05, N-14.1

**UV-VIS Spectral studies:** Four bands were showed in ethanol medium. Band A and B at 207 nm and at 268nm wavelength respectively due to transition in aromatic ring [5]. Band C appeared at 305nm wavelength due to transition in between  $\pi$  orbital localized on central azomethine (-CH=N-) bond [6]. Band D appeared at 347nm wavelength due to charge transfer within the entire Schiff base molecule this bond is commonly observed in o-hydroxyl Schiff base [7] and is based on strong intramolecular H-bonding between the hydroxyl group of the salicylidine and the azomethine nitrogen [8].

**FTIR Studies:** The absence of spikes above 3300  $\text{cm}^{-1}$  in Figure 1 show that the primary amine has formed an imines in the ligand and cannot be absorbed within that frequency range [9]. The weak absorption around 3750  $\text{cm}^{-1}$  could be attributed to-OH group. The imines  $\nu$  (C=N) functional group absorbed strongly at 1605  $\text{cm}^{-1}$ . The presence of the benzene ring conjugated to imines group in the ligand was noticed at 1551  $\text{cm}^{-1}$  [10].

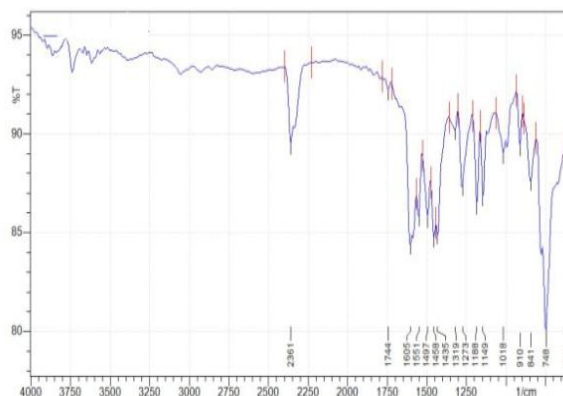


Figure-1 IR Spectra of Schiff base

**<sup>1</sup>H NMR Studies:** <sup>1</sup>H NMR data from figure 2 shows the tautomeric equilibrium favours the phenol amine in CDCl<sub>3</sub>.  $\delta=9.825$ ppm, singlet for (-CH=N-),  $\delta=6.92-7.4$  ppm for aromatic ring.

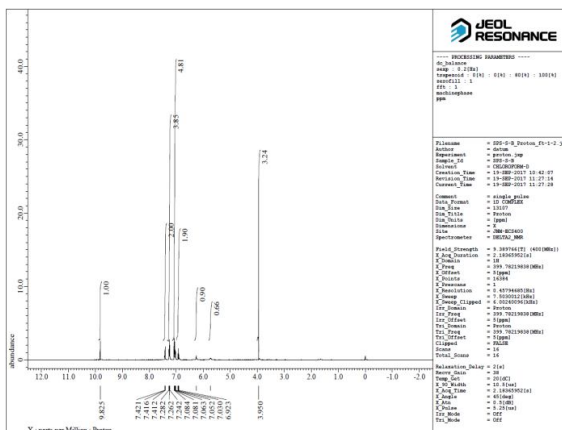


Figure 1<sup>1</sup>H NMR Spectra of Schiff base

**CHARACTERIZATION OF SILVER NANOPARTICLE:**

**UV-VIS Spectral studies:** The specific colour of colloidal silver nanoparticle solution is due to Plasmon absorbance and produces peak at 405 nm ( $\lambda_{max}$ ) wavelength. Incident light produces oscillations in conductive electrons on the surface of nanoparticles and electromagnetic radiation is absorbed so peak is observed.

**SEM Studies:** The scanning electron microscope (SEM) image shown in figure no.3. The synthesized silver nanoparticle shows the particle size is 50nm which confirms nanostructure.

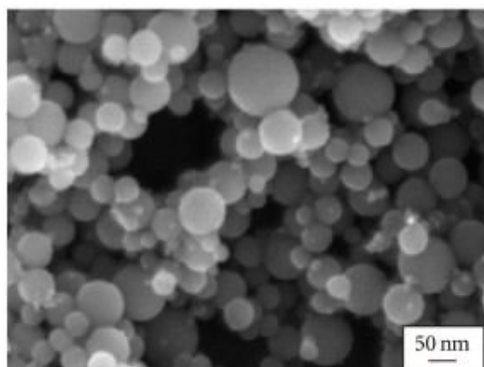


Figure-3 SEM image of silver nanoparticle

**CHARACTERIZATION OF SILVER NANOPARTICLE ANCHORED WITH SCHIFF BASE:**

**Comparison of UV-VIS Spectra of AgNps and AgNps anchored with Schiff base:** Comparative Study shown from the figure 4.

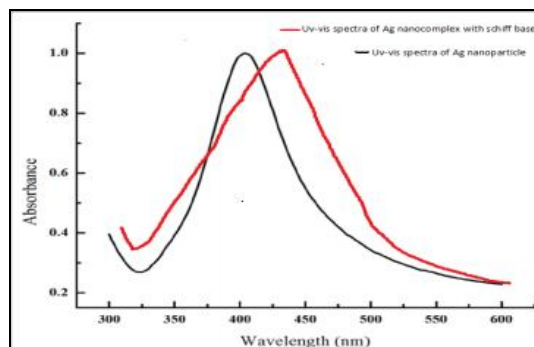


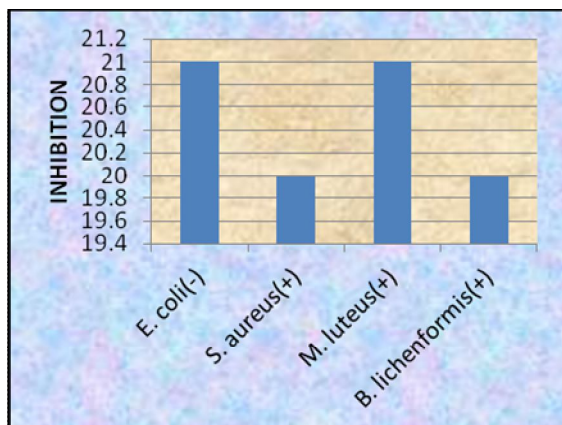
Figure 4-UV-VIS of AgNps and AgNps anchored with Schiff base

The UV-Vis spectra of silver nanoparticle (AgNps) shows  $\lambda_{max}$  at 405 nm and Schiff base complex of AgNps shows  $\lambda_{max}$  at 450 nm. A spectrum shows shifting at higher wavelength or red shift.

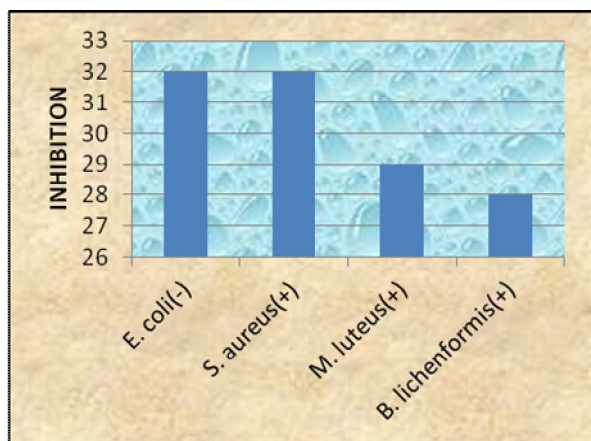
**Antibacterial Studies:** Evaluation of antimicrobial activity of all compounds *in vitro* was carried out by paper disc method against bacteria including *E. coli*, *S. aureus*, *M. luteus*, and *B. licheniformis*. Ofloxacin was additionally tested as positive control. The Disc Diffusion method [11], [12] was used to determine the antimicrobial activities of the Schiff bases using standard procedure of 6 mm disc were prepared from whatman’s filter paper no. 1. Silver nanoparticles coated Schiff base solutions of varying concentrations ranging from 100, 500, 1000 ppm was prepared. Nutrient agar was prepared, sterilized and used as the growth medium for the culture of microorganisms; 20 ml of the sterilized medium was poured into each sterilized Petri dish, covered and allowed to solidify. 16 hour old broth cultures of the specified microorganisms were used for testing antibacterial activity [13]. The sample, control and standard treated discs were air dried at room temperature, to remove any residual solvent which might interfere with the determination, sterilized and inoculated. These plates were initially placed at low temperature for 1 hour so as to allow the maximum diffusion of compounds from the test disc into the agar plate and later incubated at 37°C for 24 h in case bacteria [14], after which the zone of inhibition could be easily observed. The data represent the values of three replicates and are evaluated as mean  $\pm$  SEM values were determined and are shown in **table 2**. The Significance level of all compounds were ( $P<.001$ ), ( $*P<.01$ ). The antibacterial activity was evaluated by tube dilution method which depends on the inhibition of growth of a microbial culture in a uniform solution of antibiotic in a fluid medium that is favourable to its rapid growth in the absence of the antibiotic [15]. In this method minimum inhibitory concentration MIC of the test compounds was determined. Their MIC values in then **table 3**.

**Table-2: Antimicrobial activity of AgNps anchored with Schiff base:**

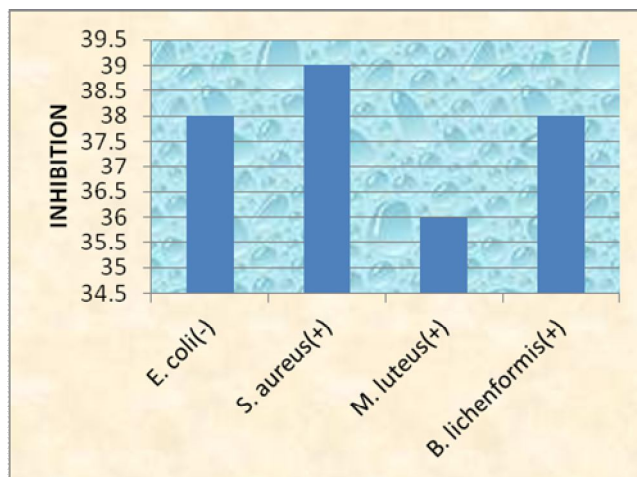
Conc.(ppm)	<i>E. coli</i> (-)	<i>S. aureus</i> (+)	<i>M. luteus</i> (+)	<i>B. lichenformis</i> (+)
100	21(±.205)	20(±.450)	21 (±.209)	20 (±.205)
500	32(±.568)	32(±.016)	29(±.805)	28(±.548)
1000	38(±.650)	39(±.360)	36(±.036)	38(±.025)



**Figure-5 Graphical representation of antibacterial activity of silver nanoparticle anchored with Schiff base at 100 ppm**



**Figure-6 Graphical representation of antibacterial activity of silver nanoparticle anchored with Schiff base at 500 ppm**



**Figure-7 Graphical representation of antibacterial activity of silver nanoparticle anchored with Schiff base at 1000 ppm**

**Table 3: MIC value (in mg/ml) of silver nanoparticle anchored with Schiff base**

<i>E. coli</i> (-)	<i>S. aureus</i> (+)	<i>M. luteus</i> (+)	<i>B. lichenformis</i> (+)
0.26	0.35	0.31	0.26

**IV. CONCLUSION**

A new silver nanoparticle anchored with Schiff bases was synthesized. The structure of synthesized compounds was confirmed on the basis of IR, NMR spectra. The Antibacterial activity of the synthesized compounds was studied using disc diffusion method and the concentration was fixed using Minimum inhibitory concentration (MIC) method. The antibacterial study revealed that all compounds showed little to excellent activity as compared to standard drug Ofloxacin.

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