

## Synthesis, Spectral Studies and Antimicrobial Activity of Zinc(II) Azide Complexes with *N,N*-Donor and *N,N,S*-Donor Schiff Base Ligands Derived from 2-Pyridinecarboxaldehyde

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Two water soluble Zn(II) Schiff base complexes of composition  $[Zn(L^1)_3][Zn(N_3)_4]$  (**1**) and  $[Zn(L^2)_2][Zn(N_3)_4]$  (**2**) were synthesized using bidentate *N,N*-donor ( $L^1$ ) and tridentate *N,N,S*-donor ( $L^2$ ) of Schiff base ligands viz. (*E*)-1-phenyl-*N*-(pyridin-2-ylmethylene)methanamine ( $L^1$ ) and (*E*)-2-((pyridin-2-ylmethylene)amino)benzenethiol ( $L^2$ ). Different spectroscopic tools such as FT-IR, UV-Vis, <sup>1</sup>H NMR and fluorescence spectroscopy have been employed to characterize complexes **1** and **2**. Efforts for getting single crystals suitable for X-ray crystal structure could not be achieved. However, from the spectroscopic data and on the basis of the structures of the related zinc(II) complexes reported earlier in the literature, the complexes could be proposed to adopt six coordinated octahedral geometries in one zinc centre coordinated to the ligands and while the other zinc centre has four azides ( $N_3$ ) terminally coordinated resulting in the octahedral and tetrahedral metal geometries around zinc metal atoms. The compounds behave as electrolytes in acetonitrile solution. *In vitro* antimicrobial activity of the Zn(II) complexes were studied against some microbes like *Aspergillus niger*, *Fusarium ventricosum* and *Fusarium chlamydosporum*. Effective antifungal activity of complex **2** was found against the tested fungal strains.

**Keywords:** Water-soluble zinc(II) azide complexes, Photophysical properties, Antimicrobial studies.

### INTRODUCTION

The aim of coordination chemistry is to search for new and versatile ligands that could be in use in many biological applications [1,2]. The combination of donor atoms (like N, S, O, etc.) in Schiff base ligands have shown a wide range of biological applications and paid particular attention over the years because of their diversity in bonding to the transition metal ions [1,2]. These donor groups acting as a significant class of ligand because of their synthetic stability toward the central metal atom [3,4]. Their worth is attributed to the presence of a Schiff base ligand containing (-N=CH-) group, which helps in elucidating the methods of many reactions in biological systems [2,5]. In addition, the heteroatom in Schiff bases (such as N, N, S) are of significant attention because of their potential applications in catalysis, engineering materials, medicine as antibiotics and anti-inflammatory agents and also in industry as corrosive resistant [6-10]. Moreover, the chemistry of transition metal complexes of ligands featuring nitrogen, oxygen and

sulfur donor atoms are very important due to the carcinostatic, antitumor, antiviral activity [11]. This type of Schiff bases and their metal complexes have shown advantages in catalyzing reactions of oxygenation of alkenes and oxidation of alcohols as well as hydrolysis and electron reduction [12-14]. It is well documented that some drugs of this type of compounds boast higher activity when administered as a metal complex rather than as free ligands [11,15-17].

In recent times, the study of transition metal complexes with N, S donor ligands have fascinated the researchers because of their encouraging antibacterial and antifungal activities [18-21]. A great deal of thought is also given to the complexes formed by 3d-metals having bidentate sulfur and nitrogen donor ligands [22,23]. The activity of these ligands and their substituted derivatives is usually improved by complexation [24-31]. Keeping all these facts into consideration, herein we report the synthesis and characterization of two new Zn(II) complexes of *N,N*-donor and tridentate *N,N,S*-donor Schiff base ligands (*E*)-1-phenyl-*N*-(pyridin-2-ylmethylene)methanamine ( $L^1$ )

and (*E*)-2-((pyridin-2-ylmethylene)amino)benzenethiol ( $L^2$ ), respectively. The spectroscopic characterizations of these complexes were studied using FT-IR,  $^1\text{H}$  NMR, UV-visible and fluorescence spectroscopies. The antimicrobial properties of the synthesized zinc complexes (**1-2**) were studied against some microbial species like *Aspergillus niger*, *Fusarium ventricosum* and *Fusarium chlamydosporum*.

## EXPERIMENTAL

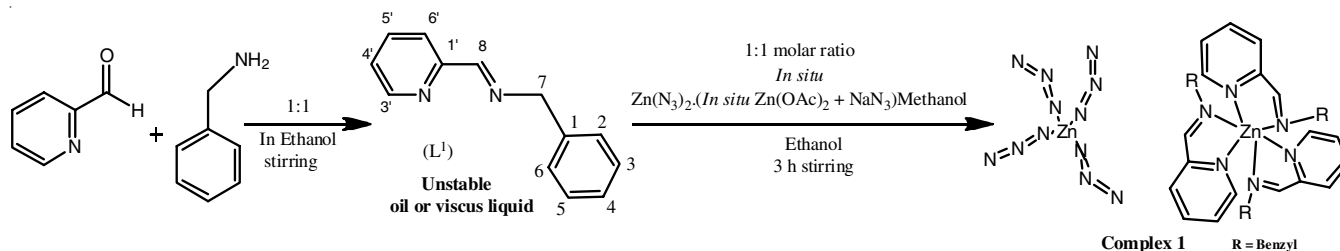
2-Pyridinecarboxaldehyde (Avra Pvt. Ltd.), benzylamine (Sigma-Aldrich), 2-aminothiophenol (Merck),  $\text{Zn}(\text{OAc})_2 \cdot 2\text{H}_2\text{O}$  (Merck), sodium azide (Sigma-Aldrich) were purchased and used without further purification. Purification of the solvents was carried out by normal procedures. Scanca apparatus was used to record the melting point of compounds by capillary tube and are uncorrected. The Shimadzu FT-IR-8400S spectrophotometer were used to record the IR spectra of the complexes using KBr pellets in the range of  $4000\text{--}400\text{ cm}^{-1}$  and UV-1800 Shimadzu spectrophotometer were used to record UV-visible spectra of the complexes in acetonitrile solution at  $200\text{--}800\text{ nm}$  range. The fluorescence spectra of the complexes were recorded and corrected in instrument response function on a Perkin-Elmer spectrofluorimeter model LS55 (with 10 and 15 nm width fixed slits for excitation and emission). The photophysical properties were measured using 10 mm quartz cuvettes (Perkin-Elmer B0831009) optical path length for absorption and for fluorescence, Hellma Germany (type 111-QS) were used, respectively. Quantum yield ( $\phi_F$ ) for fluorescence of the complexes were calculated using a standard which has been described earlier [32]. Experimental error of the measured  $\phi_F$  with that of the instrument was taken to be within  $\pm 10\%$ . Electrical conductivity was measured using eutech precision bridge Con510 automatic instrument analyser. Bruker Avance II spectrometer was used to measure  $^1\text{H}$  NMR spectra at  $400.13\text{ MHz}$  and chemical shifts were recorded using  $\text{Me}_4\text{Si}$  set as a reference at  $0.00\text{ ppm}$ .

**Synthesis of ligands  $L^1$  and  $L^2$ :** *In situ* reactions a pale yellow liquid of ligand  $L^1$  reported earlier [33] and followed the same procedure. Ligand  $L^2$  generated *in situ* reaction of pyridine-2-aldehyde and 2-aminothiophenol in ethanol solution.

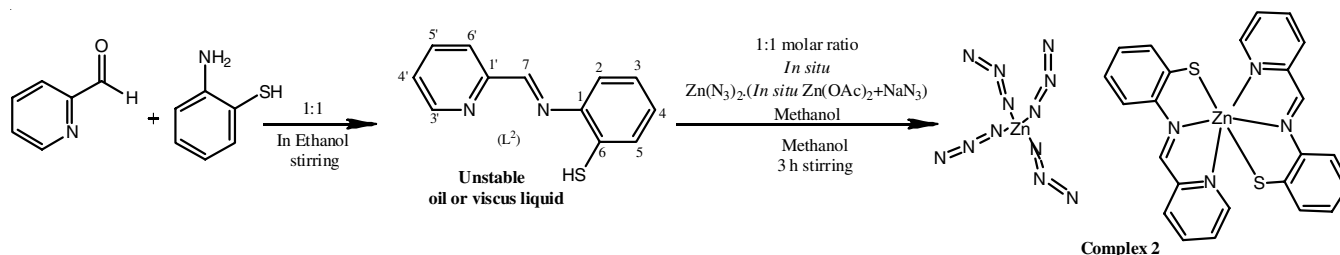
## Synthesis of zinc(II) complexes

**Synthesis of [((*E*)-1-phenyl-*N*-(pyridin-2-ylmethylene)-methanamine) $_3\text{Zn}(\text{II})][\text{Zn}(\text{N}_3)_4]$  (**1**):** *In situ* reactions, in ethanol equimolar amount of benzylamine (0.15 g, 1.40 mmol) was added to 2-pyridine carboxaldehyde (0.15 g, 1.40 mmol). The reaction mixture was stirred for 20 min and then  $\text{Zn}(\text{N}_3)_2$  was added slowly under stirring condition (prepared *in situ* with the reaction between  $\text{Zn}(\text{OAc})_2 \cdot 2\text{H}_2\text{O}$  (0.256 g, 1.40 mmol) and  $\text{NaN}_3$  (0.18 g, 2.77 mmol) in methanol), which resulted in the instant formation of a pale yellow precipitate. The stirring was continued for 3 h and then the mixture was filtered. The residue was collected and dried in a vacuum (**Scheme-I**). Yield: 71%. m.p.  $149\text{--}150\text{ }^\circ\text{C}$ . Anal. calcd. (found) % for  $\text{C}_{39}\text{H}_{36}\text{N}_6\text{Zn}\cdot\text{N}_{12}\text{Zn}$ : C, 52.77 (51.90); H, 4.05 (3.90); N, 28.41 (28.23).  $\Lambda_m$  ( $\text{CH}_3\text{CN}$ ):  $150\ \Omega^{-1}\text{ cm}^2\text{ mol}^{-1}$ . IR (KBr,  $\text{cm}^{-1}$ ):  $1605\ \nu_{\text{asym}}(\text{C}(\text{H})=\text{N})$ ; 1508, 1475, 1395  $\nu(\text{C}=\text{N})_{\text{py}}$ ; 2065ss:  $\nu(\text{N}_3)$ ;  $^1\text{H}$  NMR ( $\text{DMSO-}d_6$ ):  $\delta$  8.57 (d, 1H, H-3'), 8.51 (s, 1H, H-8), 8.14 (t, 1H, H-42), 7.93 (d, 1H, H-62), 7.63 (t, 1H, H-52), 7.23 (d, 2H, H-2,6), 7.02 (b, 3H, H-3,4,5), 4.77 (s, 2H, H-7) ppm.

**Synthesis of [((*E*)-2-((pyridin-2-ylmethylene)amino)benzenethiol) $_3\text{Zn}(\text{II})][\text{Zn}(\text{N}_3)_4]$  (**2**):** Compound **2** was synthesized following the analogous procedure as in case of **1** except that benzylamine was used instead of 2-aminothiophenol (**Scheme-II**). Yield: 63%. m.p.  $171\text{--}172\text{ }^\circ\text{C}$ . Anal. calcd. (found) % for  $\text{C}_{24}\text{H}_{18}\text{N}_4\text{S}_2\text{Zn}\cdot\text{N}_{12}\text{Zn}$ : C, 39.75 (39.00); H, 2.50 (2.10); N, 30.90 (30.00).  $\Lambda_m$  ( $\text{CH}_3\text{CN}$ ):  $156\ \Omega^{-1}\text{ cm}^2\text{ mol}^{-1}$ . IR (KBr,  $\text{cm}^{-1}$ ):  $1620\ \nu_{\text{asym}}(\text{C}(\text{H})=\text{N})$ ; 1585, 1500, 1470  $\nu(\text{C}=\text{N})_{\text{py}}$ ; 2050ss:  $\nu(\text{N}_3)$ .  $^1\text{H}$  NMR ( $\text{DMSO-}d_6$ ):  $\delta$  9.17 (s, 1H, H-7), 8.70 (d, 1H, H-32), 8.24 (t, 1H, H-42), 8.00 (d, 1H, H-62), 7.80 (t, 1H, H-52), 7.61 (d, 1H, H-2), 7.40 (d, 1H, H-5), 57.05 (t, 1H, H-3), 6.90 (t, 1H, 4) ppm.



**Scheme-I:** Synthesis for the complexes **1**



**Scheme-II:** Synthesis for the complexes **2**

**Antimicrobial assay:** The antimicrobial screening test was performed by modified Kirby-Bauer disc diffusion assay [34]. Three bacteria (*Escherichia coli*, *Pseudomonas sp.* and *Staphylococcus aureus*) and three fungi (*Aspergillus niger*, *F. chlamydosporum* and *Fusarium ventricosum*) were used for the microbial growth inhibition assay. Prior to assay, all the organisms were revived by streaking on agar media. For preparing bacterial inoculums, fresh cultures were inoculated in nutrient broth medium and incubated for 24 h at 37 °C and subsequently diluted up to 1:4 ratio with water. While for preparing fungal inoculums, 7 days old fungal culture were used. A sterile distilled water (5 mL) was added to the plates containing the fungal cultures and the colonies were gently scraped out along with water. The fungal spore and hyphal suspension was vortexed at a minimum speed for 30 s to 1 min and used as inoculums.

The filter paper discs (6 mm diameter discs prepared from Whatman No.1) were sterilized by autoclaving prior to the assay. A thin layer of bacterial inoculum (150  $\mu$ L of bacterial cell suspension, 1 mg mL<sup>-1</sup>) was prepared using an L-spreader on the surface of Tomato Juice Agar (TJA) plates for bacteria. Similarly, fungal inoculum (150  $\mu$ L of 1 mg mL<sup>-1</sup> and 2 mg mL<sup>-1</sup>) was inoculated on the surface of Rojo Congo Agar (RCA) plates and spread uniformly with an L-spreader. The plates were allowed to dry for about 1 min inside the laminar airflow chamber. Then, 3 filter paper discs previously saturated with test sample solutions (1 mg mL<sup>-1</sup> for bacteria) and (1 mg mL<sup>-1</sup> and 2 mg mL<sup>-1</sup>) for fungi were placed on the surface of the plated inoculated with bacteria and fungi. A standard antibiotic disc of streptomycin (1 mg mL<sup>-1</sup>) for bacteria and amphotericin B (1 mg mL<sup>-1</sup> and 2 mg mL<sup>-1</sup>) for fungi were also included for each of the organisms tested. The plates were incubated for 24 h at 37 °C for bacteria and 72 h at 30 °C for fungi. The growth inhibition zones on the bacterial and fungal plated were recorded for the samples as and the standard antibiotics. The entire assay was performed in triplicate plates for all the samples and antibiotics.

## RESULTS AND DISCUSSION

In alcohol *in situ* reactions, the equimolar amount of Zn(N<sub>3</sub>)<sub>2</sub> (generated *in situ* from Zn(OAc)<sub>2</sub>·2H<sub>2</sub>O and NaN<sub>3</sub>) reacted rapidly with L<sup>1-2</sup> [generated *in situ* from the condensation reaction of pyridine-2-carboxaldehyde and benzylamine (L<sup>1</sup>) or 2-aminothiophenol (L<sup>2</sup>)] to give a pale yellow precipitate of Schiff base metal complexes **1-2**, similar to that reported for analogues ligands [33-35]. The general stoichiometries of the complexes, [Zn(L<sup>1</sup>)<sub>3</sub>][Zn(N<sub>3</sub>)<sub>4</sub>] (**1**) and [Zn(L<sup>2</sup>)<sub>2</sub>][Zn(N<sub>3</sub>)<sub>4</sub>] (**2**) have been compared by the microanalytical results, which clearly evidenced the formation of 3:2 adducts between the bidentate *N,N*-donors ligand (L<sup>1</sup>) with Zn(N<sub>3</sub>)<sub>2</sub> for complex **1** and formation of 2:2 adduct between tridentate *N,N,S*-donor ligands (L<sup>2</sup>) with Zn(N<sub>3</sub>)<sub>2</sub> for complex **2**. The complexes are air-stable and were obtained in moderate to good yields. The complexes behave as electrolytes in acetonitrile solution.

**IR spectral studies:** The infrared spectra of complexes **1** and **2** are similar. The effect of coordination of ligand to zinc(II) upon the IR spectrum could not be assigned, in the absence of the IR data for the free ligands. The complexes displayed a

moderately intense IR band in the region 1620-1590 cm<sup>-1</sup>, which is assigned to be  $\nu_{\text{asym}}(\text{C(H)=N})$  stretching of the coordinated Schiff base ligands [35-37]. In addition, a well-resolved sharp bands of variable intensity observed in the regions 1600-1580, 1490-1475 and 1450-1435 cm<sup>-1</sup> are assigned to the coordinated pyridine ring [35-37]. The complexes showed a very strong band at approximately 2060-2030 cm<sup>-1</sup> suggesting the presence of  $\nu_{\text{asym}}(\text{N}_3)$ (azide) [35-39]. In addition, a strong band observed at around 2050 cm<sup>-1</sup>, indicates the presence of non-bridge azide [40-42].

**<sup>1</sup>H NMR spectral studies:** The <sup>1</sup>H NMR spectra of the synthesized zinc complexes (**1-2**) were obtained in DMSO-*d*<sub>6</sub> solution and displayed the estimated signals which correlate well with the hydrogen atoms present in the molecules and therefore, relates the presence of the ligand skeleton in the respective complexes [42-44]. The <sup>1</sup>H NMR chemical shifts for the synthesized complexes are based on the splitting patterns of the signals by comparing the data with those in previous information [35,36,45]. The effect of coordination to zinc(II) upon the <sup>1</sup>H NMR chemical shifts could not be assigned, in the absence of the NMR data for the free ligands, which could only be prepared *in situ*. In both complexes, a singlet peak was observed at around 9.20-8.51 ppm which can be assigned for Schiff base (C(H)=N) protons [36,44]. The doublet and triplet signals observed at *ca.* 8.26-6.52 ppm can be assigned to *pyridine* rings [43]. The <sup>1</sup>H NMR spectrum of complex **1** shows a singlet peak at *ca.* 4.77 ppm, which can be assigned to benzyl-CH<sub>2</sub> protons. In complex **2** the absence of -SH protons, indicates the coordination of sulphur moiety to the metal. Aromatic protons were observed as singlet, doublet and triplet in the complexes in the range 6.03 to 8.10 ppm.

**Conductance:** Molar conductance values of the synthesized zinc complexes were range from 150-156  $\Omega^{-1}$  cm<sup>2</sup> mol<sup>-1</sup>, which show that the complexes are electrolytic in nature in acetonitrile solution [43].

**UV-visible and fluorescence spectral studies:** The UV-visible and fluorescence spectra were recorded in the acetonitrile solution at a concentration of 10<sup>-5</sup> M. The electronic properties of the synthesized zinc complexes **1-2** are shown in Table-1. The spectra of the complexes remain unchanged over a period of one week. The absorption spectra of the complexes were recorded in the range of 200-800 nm. Both zinc complexes **1-2** commonly consists of one intense band (Fig. 1a). The electronic spectra of the ligands are not available [35,44]. Broad fluorescence emission was observed for both complexes at  $\lambda_{\text{em}} = 375$ -450 nm (Fig. 1b). Even though, these broad fluorescence emissions could not be confined as a charge transfer transition of metal-to-ligand (M→L) or ligand-to-metal (M←L). The available zinc(II) complexes, with a *d*<sup>10</sup> arrangement reported earlier, shown that the complexes not easily oxidized or reduced [46]. Nevertheless, the synthesized zinc complexes **1-2** were attributed to intraligand (IL) ( $\pi$ - $\pi^*$ ) transition [35,36,43]. The complexes show very low fluorescence quantum yield [47,48] and found to be very weak emitter at room temperature.

**Proposed structure of water soluble zinc(II) complexes:** Although a very few amount of work has been reported with similar ligands environment. To the best of our knowledge the

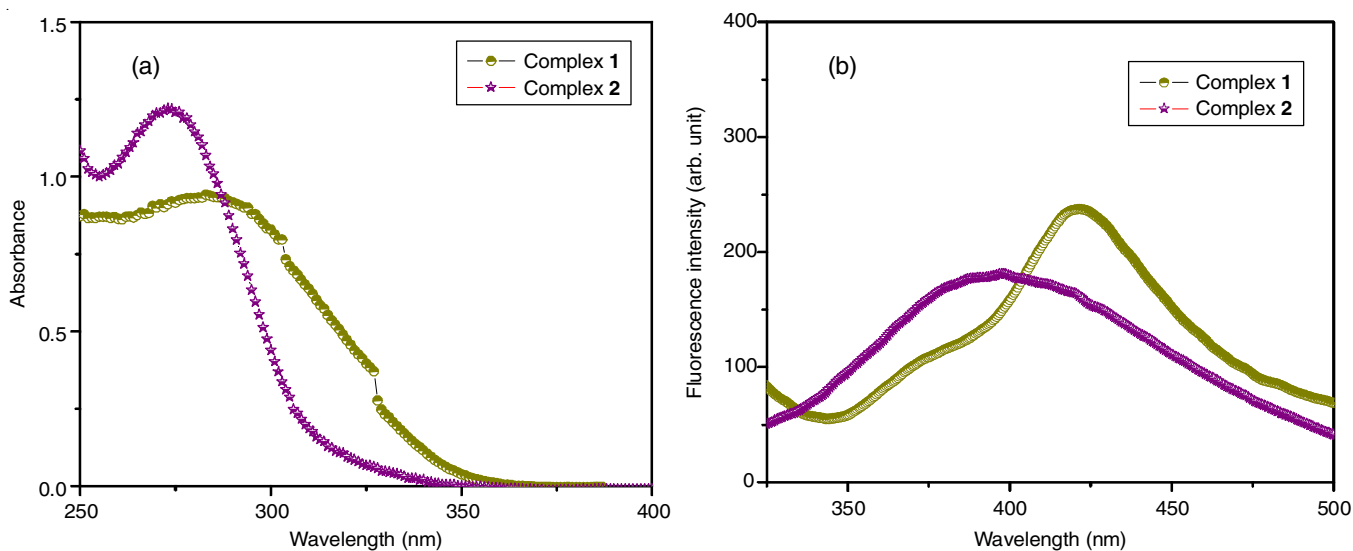


Fig. 1. UV-Vis spectra (a) and fluorescence spectra (b) of complexes were obtained by the excitation of absorption maxima respectively in acetonitrile solution at concentration  $\sim 10^{-5}$  M

TABLE-1  
UV-VISIBLE AND FLUORESCENCE DATA IN  
ACETONITRILE SOLUTION FOR COMPLEXES 1-2

Complexes	UV-Visible	Fluorescence data	
	$\lambda_{\text{max}}$ (nm)	$\lambda_{\text{em}}$ (nm)	$\phi_{\text{F}}$
1	280	420	0.09
2	275	404	0.11

existing literature holds no crystallographic characterization of the ligands except for the aniline derivatives [49]. Previously, Basu Baul *et al.* [50] carried out a quantum chemical calculations for the related ligands to examine infrared bands and compared with the experimental values. The crystal structures of few analogous ligands with mercury(II) azide and zinc(II) azide complexes were also reported [35,36,40,43,44]. Nevertheless, in the coordination chemistry, zinc(II) ions with *N,N*-donor and *N,N,S*-donor ligand atmosphere, typically prefer to have a four coordinated tetrahedral complex and also depending on the bulkiness of the ligands it prefers to form five and six coordinated trigonal bipyramidal and octahedral complexes [40,50,51-54]. However, from the spectroscopic analysis of the similar ligand system reported earlier [40,43,55,56], the ligands are proposed to be bidentate and tridentate nature of coordination to the metal. The IR spectra of complex 1 indicated that  $L^1$  behaves as a bidentate ligand, coordinated to the metal ions *viz.* azomethine N, imine N while in complex 2,  $L^2$  behaves as a tridentate ligand, coordinated to the metal ions *viz.* azomethine N-, imine N- and S- moiety of 2-aminothiophenol. In these zinc complexes, it is anticipated that the azide ion is coordinated to the metal as non-bridging terminal fashion and it is reflected in broad unresolved IR absorption band at around  $2037-86 \text{ cm}^{-1}$  [39,40,42,57,58].

From the molar conductance data, it was found that the Zn(II) chelates may be considered as electrolytes in solution [40,43]. The  $^1\text{H}$  NMR spectra of the complexes are in good agreement with the formation of the expected compounds [35,43]. The results of the crystal structures and spectroscopic

studies of *N,N*-donor and *N,N,S*-donor ligand systems with Zn(II) ion reported earlier [33,40,50,51-54,56] are consistent with the spectroscopic properties of synthesized zinc complexes (1-2). Complex 1 assumed to be 2:3 adduct between zinc(II) azide and ligand ( $L^1$ ) [33,57] and in complex 2, assumed to be 2:2 adduct between zinc(II) azide and ligand ( $L^2$ ). Moreover, in both complexes (1-2), there are two zinc centres [33,56,57]. The proposed octahedral and tetrahedral structures in two zinc centre of the complexes (1-2), is based on the spectroscopic data of the complex and reported structure [33]. In both synthesized zinc complexes (1-2), four azides are predicted to be present as non-bridging terminal bonding to the one metal centre [59] and other metal centre coordinated to *N,N*-donor atom of the three ligands in complex 1 [33,39,54,56,59,60] and in complex 2, other metal centre coordinated to N, N, S donor atom of two ligands [56,57,60-65]. The proposed octahedral structure of the complexes, is based on when the azides are assumed to be present as non-bridging terminal [40,42,57,59] bonding and it is also supported from the X-ray crystal structure of Zn(II), two metal centre non-bridging complexes [33,54,56,60,62,66,67]. The similar ionic form of the complexes with two zinc centre and zinc-containing four-terminal chloride ions are also reported [33,59]. In short, the proposed octahedral and tetrahedral structure of two zinc centre of Zn(II) azide complexes are tentative and difficult to assign a conclusive structure without single crystal X-ray structure.

**Antimicrobial activity:** The synthesized zinc complexes 1 and 2 were screened for their antimicrobial activity against three bacteria and three fungal species. There was no inhibition of growth by compound 1 and 2 against the three bacteria tested. Streptomycin showed inhibition of growth in all the plates inoculated with *S. aureus*. For fungal plates, at  $2 \text{ mg mL}^{-1}$  complex 2 showed an inhibition zone of 0.47 mm on plates inoculated with *A. niger* and the results are shown in Fig. 2a. There was no growth inhibition properties of complex 2 against *F. ventricossum* and *F. conidiosporum*. Complex 1 showed no inhibition activity against all the three fungi assayed in the



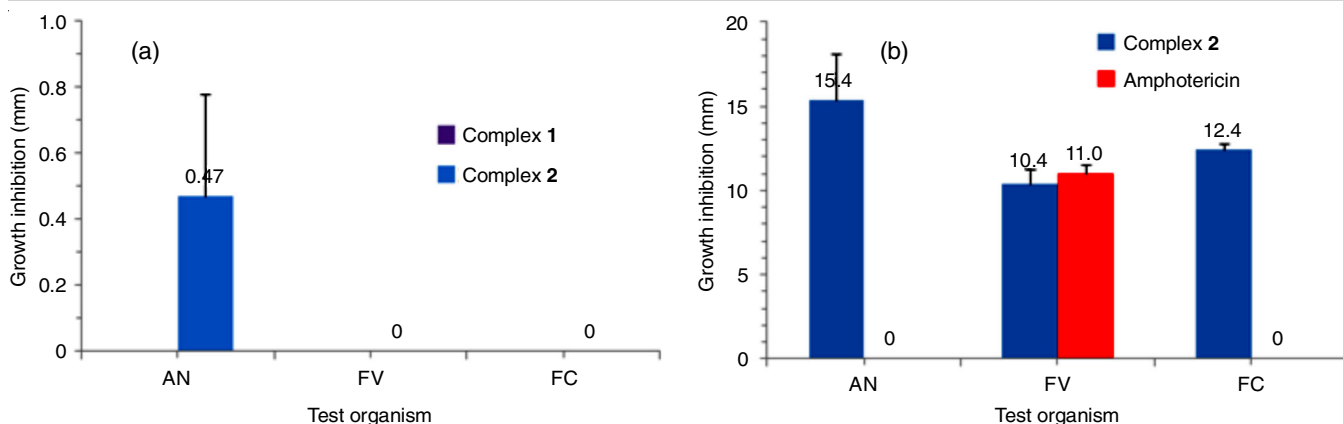


Fig. 2. (a) Antimicrobial properties of **1** and **2** on three fungi [*Aspergillus niger* (AN), *Fusarium ventricosum* (FV) and *Fusarium chlamydosporum* (FC)] at a concentration of 1 mg mL<sup>-1</sup>. (b) Antimicrobial properties of **2** on three fungi (AN, FV and FC) at a concentration of 2 mg mL<sup>-1</sup>. Amphotericin (AMP) was included as standard microbial growth inhibitor. T-bars on the histogram represent standard deviation ( $\pm$ ) of the triplicate values

experiment. Complex **2** exhibited inhibition of growth at 2 mg mL<sup>-1</sup> against three fungi when the concentration was increased from 1 mg mL<sup>-1</sup> to 2 mg mL<sup>-1</sup>. The maximum inhibition of 15.4 mm was observed against *A. niger* followed by 12.4 mm for *F. conidosporum* as shown in Fig. 2b. A minimum inhibition of 10.4 mm of complex **2** was recorded against in *F. ventricosum*. Thus, complex **2** showed antifungal activity, however, it does not exhibit any antibacterial activity against the tested bacteria.

### Conclusion

In summary, the synthesis of two water soluble zinc(II) azide complexes with ligands *viz.* (*E*)-1-phenyl-*N*-(pyridin-2-ylmethylene)methanamine(**L**<sup>1</sup>) and (*E*)-2-((pyridin-2-ylmethylene)amino)benzenethiol (**L**<sup>2</sup>) were successfully accomplished. The synthesized complexes have been characterized by FT-IR, <sup>1</sup>H NMR, UV-visible and fluorescence spectroscopy. From the spectral studies, the complexes showed that both ligands (**L**<sup>1-2</sup>) are coordinated to the zinc metal. Based on the spectroscopic studies, complex **1** form octahedral and tetrahedral metal geometries around two Zn(II) metal atom. Whereas complex **2** suggested to be six coordinated octahedral geometry and tetrahedral metal geometries around two centers Zn(II) metal atom. The photophysical properties of the complexes show  $\pi$ - $\pi^*$  (intra-ligand) transition and Zn compounds show weak emitters at room temperature. Both synthesized zinc(II) complexes were also studied for their antimicrobial activities against some microbes and compared with standard drugs. Complex **2** was found to exhibit effective antifungal activity against the three tested fungal strains.

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### CONFLICT OF INTEREST

The authors declare that there is no conflict of interests regarding the publication of this article.

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