Biointerface Research in Applied Chemistry
Platinum Open Access Journal (ISSN: 2069-5837)

Article
Volume 12, Issue 3, 2022, 3607 - 3617
https://doi.org/10.33263/BRIAC123.36073617

Transition Metal (II) Complexes of (*E*)-*N*-(4-methylbenzylidene)-2-((*Z*)-(4-methylbenzylidene)amino)benzamides: Synthesis, Characterization and their Biological Evaluation

Venkatesh K Bhovi [1,*], Bharathi K [1], Sulochana P Melinmath [1], Vrushabendra Basavanna [2], Srikantamurthy Ningaiah [2,*]

[1] P G Studies and research in Chemistry, Vijayanagar College Hosapete, Vijayanagar, Karnataka, India -583201
[2] Department of Chemistry, Vidyavardhaka College of Engineering, Visvesvaraya Technological University, Mysuru-570 002, Karnataka, India
* Correspondence:venki.kb123@gmail.com (V.K.B.);srijmn@vvce.ac.in(S.N.)

Scopus Author ID35096095900;55786395000
Received: 31.05.2021; Revised: 5.07.2021; Accepted: 9.07.2021; Published: 10.08.2021

Abstract: A novel series of transition metal (II) complexes (5a-h) were conveniently synthesized via reaction of important transition metals (Co, Cu, Zn, Ni) with (E)-N-(4-methylbenzylidene)-2-((Z)-(4-methylbenzylidene)amino)benzamide Schiff base (3) which was previously synthesized by reacting 2-aminobenzohydrazide (1) with 4-methylbenzaldehyde (2). The synthesized metal complexes' structure was elucidated by IR, NMR, mass, and elemental analysis. Additionally, we also evaluated the antioxidant, antimicrobial and antifungal activity of the synthesized metal complexes. The bioassay of the novel transition metal complexes envisioned that compounds5eand5c showed better antimicrobial activity than the free ligand, and compounds5g and 5a showed good activity against most bacterial strains. On the other hand, hydrated metal complexes 5b, 5d, 5f, and 5h showed moderate to good antimicrobial activity. In comparison with ascorbic acid, most of the metal complexes showed moderate to good antioxidant activity. The current bioassay was investigated and proved that the compounds 5e and 5c as antimicrobial agents act on highly resistant strains of microbes.

Keywords: metal (II) complex; Schiff's base; antimicrobial; ascorbic acid.

1. Introduction

Schiff's base is a sub-class of organic imines considered secondary aldimines or ketimines, depending upon the structure [1]. A new era has started in coordination chemistry since 1869 after Schiff's elegant synthesis of azomethane complexes of copper(II) from preformed metal, salicylaldehyde, and primary amine [2]. Schiff base is formed as a condensation product of primary amine with carbonyl compounds [3]. This was first reported by Schiff [4], which contains the >C=N- group, which is also called azomethine or imine. The >C=N- group, combined with more such groups or others like phenolic –OH or amino groups, can effectively form metal complexes. The Schiff-based metal complexes have shown evidence of importance significant in inorganic and organic chemistry due to their biological activity. In recent years several reports are published on the preparation of these compounds and their application [5]. Schiff bases of aliphatic aldehydes are comparatively unstable and readily undergo polymerization, whereas aromatic aldehydes possessing effective conjugation are

stable. In general, aldehyde compounds react faster than ketones in condensation reactions and readily yield Schiff bases as the reaction center of aldehyde is sterically less hindered than ketone [6]. Furthermore, the extra carbon of ketone compounds donates electron density to the azomethine carbon and thus makes the ketone less electrophilic compared to aldehyde [7].

The reactivity of aldehyde towards electrophilic reactions attracts the chemist to synthesize novel Schiff's base, which acts as a ligand by treating with a suitable amine or acid hydrazide. Moreover, Schiff bases have been reported to exhibit a wide variety of biological actions under the azomethine linkage, which is responsible for various antibacterial [8], antifungal [9], antitumor [10], herbicidal [11], and antioxidant [12-13] activity. These compounds were also synthesized to test their inhibition activity against α-glucosidase [14]. Similarly, metal complexes procured from Schiff's base and metal salts and screened for their antifungal [15-16], antimicrobial [17], antitumor [18], antioxidant [19-20], antimalarial [21], cytotoxicity [22] activity. These complexes were also tested for the inhibition of human phosphatases [23], cleavage efficiency against pBR322 DNA [24], and for the treatment of hepatocellular carcinoma [25]. The above study on a wide variety of applications of Schiff's base and its metal complexes in the biological field motivated us to synthesize the transition metal (II) complexes through Schiff's base intermediate.

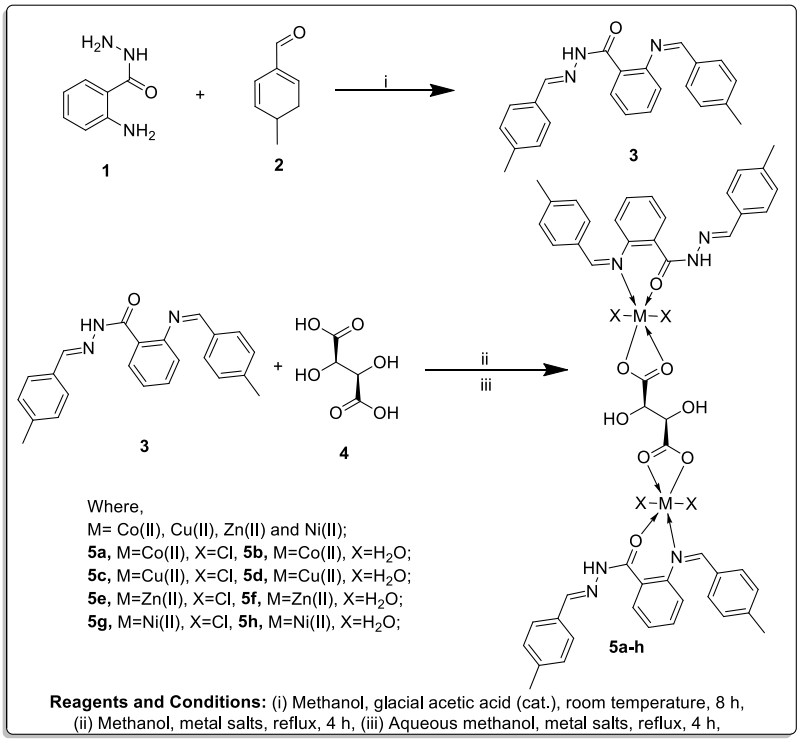
2. Materials and Methods

2.1. Chemistry.

The present research work focuses on the synthesis of ligands and their transition metal complexes. Also, the details of various physicochemical techniques employed to characterize parent ligands, corresponding complexes, and the experimental protocols followed for the evaluation of biological activity are summarized in detail.

The 4-methylbenzaldehyde and 2-aminobenzohydrazide were purchased from S.D. fine chemical India. The tartaric acid, methanol, and glacial acetic acid were procured from Aldrich, India. All the purchased chemicals were of analytical grade and used as supplied without further purification. Spectral grade deuterated DMSO-d_6 and $CDCl_3$ were used to record the NMR spectra. HPLC grade methanol and acetonitrile were used to record LCMS spectra of the ligand as well as transition metal complexes. Metal chlorides used to prepare complexes in the present work are $CoCl_2.6H_2O$, $CuCl_2.2H_2O$, anhydrous $ZnCl_2$and $NiCl_2.6H_2O$. These metal salts were procured from S.D. fine and Spectrochem Chemicals, India. Solvents were purchased from S.D. fine; most of the time, new bottles were taken for ligand and metal complex synthesis. The drying reagents employed at various stages viz., anhydrous sodium sulfate, anhydrous magnesium sulfate, anhydrous calcium chloride, and mineral acids such as hydrochloric acid, sulphuric acid, nitric acid, and bases like sodium hydroxide pellets and ammonia were of analytical grade obtained from S.D. fine Chemicals, India.

Our aim of the experiment is to synthesis and biological studies of mixed ligand metal complexes in two steps by taking two different ligands such as (*E*)-*N'*-(4-methylbenzyldene) 2-[(*E*)-(4-methylbenzylidene)amino)benzolhydrazide)] (**3**) and tartaric acid (**4**). Literature reveals that nitrogen-containing heterocyclic compounds showed good biological activities. The reaction involves two steps, step 1; the synthesis of Schiff base by taking 4-methylbenzaldehyde and 2-amino-benzohydrazide in the ratio of 2:1, and the second step involves the preparation of metal complexes by taking Schiff base, tartaric acid the metal chloride in the ratio of 2:1:2 equivalent respectively as shown in the Scheme 1.



Scheme 1. Synthesis of novel transition metal (II) complexes (**5a-h**).

2.1.1. Typical procedure for synthesis (*E*)-N-(4-methylbenzylidene)-2-((*Z*)-(4-methylbenzylidene)amino)benzamide Schiff's base(3).

The methanolic mixture (20 ml) of 2- aminobenzohydrazide(**1**, 1.51g, 10.00 mmol) and 4-methylbenzaldehyde(**2,**2.40 g, 20.00mmol)was taken in a 50 ml round bottom flask fitted with a water cooled condenser and stirred for 2-3 hours. During the course of the reaction 2-3 drops of acetic acid (catalytic amount)was added. After completion, the solid separated were filtered, washed repeatedly with cold ethanol and dried in air. The purity of the compound **(3)**was checked by TLC on pre-coated silica gel plate.The crude compound was recrystallized from methanol to procure (*E*)-*N*-(4-methylbenzylidene)-2-((*Z*)-(4-methylbenzylidene) amino) benzamide Schiff base (3.07 g) as white solid in 90.2% yield. m.p: 289 ^{o}C. IR (Nujol): 3737 cm^{-1} (N-H), 3271 cm^{-1} (C-H), 3270 cm^{-1} (O-H), 1656 cm^{-1} (C=O), 1610 cm^{-1} (C=N), 1144 cm^{-1} (C-O); ^{1}H NMR (400 MHz, $CDCl_3$): δ 2.34 (s, 6H, -CH_3), 7.28-7.29 (d, 4H, ArH), 7.64-7.91 (m, 8H, ArH), 8.03 (s, 1H, NH), 8.18 (s, 1H, =CH-), 8.79 (s, 1H, =CH-); ^{13}C NMR (400 MHz, $CDCl_3$): δ20.8, 126.4, 128.7, 129.2, 129.9, 130.6, 131.6, 133.1, 134.6, 141.6, 154.1, 159.9, 164.2, 175.8; MS (relative abundance) m/z = 356.10 $[M+H]^{+}$; Anal. % Calc for $C_{23}H_{21}N_3O$: C 77.72, H 5.96, N 11.82, O 4.50; Found: C 77.69, H 5.99, N 11.80, O 4.52.

2.1.2. Typical procedure for the synthesis of transition metal complexes (5a-h).

The ligand (*E*)-*N*-(4-methylbenzylidene)-2-((*Z*)-(4-methylbenzylidene) amino) benzamide (**3**, 0.356 g1.00 mmol) and tartaric acid(0.075 g,0.50 mmol) were stirred initially with transition metal(II) chlorides in 2:1:1 molar ratio in 20 ml of dry methanol/aqueous methanol solution, and then the reaction mixtures were refluxed on a water bath at 80 ^{o}C for over 3 hours. The color of the reaction mixtures was discharged during the reaction completion. After completion of the reaction, the resultant complexes were filtered, washed with methanol, and then with chloroform and air-dried to afford series of transition metal complexes (**5a-h**).

Cobalt metal complex (5a)

Obtainedfrom(E)-N-(4-methylbenzylidene)-2-((Z)-(4-methylbenzylidene)amino)benzamide (3, 0.356 g 1.00 mmol), tartaric acid(0.075 g,0.50 mmol), cobalt (II) chloride hexahydrate (0.237 g, 1.00 mmol) in dry methanol (20 ml) as light pink coloured solid (0.79 g) in 70.7% yield. IR (Nujol): 3742 cm^{-1} (N-H), 3270 cm^{-1} (O-H), 3269 cm^{-1} (C-H), 1650 cm^{-1} (C=O), 1613 cm^{-1} (C=N), 1132 cm^{-1} (C-O); MS (relative abundance) m/z = 1119.0 $[M+H]^+$; Anal. % Calc for $C_{50}H_{46}Cl_4Co_2N_6O_8$: C 53.69, H 4.14, Cl 12.68, Co 10.54, N 7.51, O 11.44; Found: C 53.63, H 4.19, Cl 12.68, Co 10.52, N 7.51, O 11.47.

Cobalt metal complex (5b)

Obtained from(E)-N-(4-methylbenzylidene)-2-((Z)-(4-methylbenzylidene) amino) benzamide (3, 0.356 g 1.00 mmol), tartaric acid(0.075 g,0.50 mmol), cobalt (II) chloride hexahydrate (0.237 g, 1.00 mmol) in aqueous methanol (20 ml) as dark pink coloured solid (0.69 g) in 69.6% yield. IR (Nujol): 3737 cm^{-1} (N-H), 3276 cm^{-1} (O-H), 3265 cm^{-1} (C-H), 1661 cm^{-1} (C=O), 1621 cm^{-1} (C=N), 1145 cm^{-1} (C-O); MS (relative abundance) m/z = 1050.0 $[M+H]^+$; Anal. % Calc for $C_{50}H_{54}Co_2N_6O_{12}$: C 57.26, H 5.19, Co 11.24, N 8.01, O 18.30; Found: C 57.20, H 5.20, Co 11.27, N 8.00, O 18.33.

Copper metal complex (5c)

Obtained from(E)-N-(4-methylbenzylidene)-2-((Z)-(4-methylbenzylidene) amino) benzamide (3, 0.356 g 1.00 mmol), tartaric acid(0.075 g,0.50 mmol), copper (II) chloride dihydrate (0.170 g, 1.00 mmol) in dry methanol (20 ml) as light red coloured solid (0.71 g) in 71.9% yield. IR (Nujol): 3749 cm^{-1} (N-H), 3276 cm^{-1} (O-H), 3263 cm^{-1} (C-H), 1659 cm^{-1} (C=O), 1618 cm^{-1} (C=N), 1127 cm^{-1} (C-O); MS (relative abundance) m/z = 1128.0 $[M+H]^+$; Anal. % Calc for $C_{50}H_{46}Cl_4Cu_2N_6O_8$: C 53.25, H 4.11, Cl 12.57, Cu 11.27, N 7.45, O 11.35; Found: C 53.15, H 4.19, Cl 12.53, Cu 11.30, N 7.46, O 11.36.

Copper metal complex (5d)

Obtained from(E)-N-(4-methylbenzylidene)-2-((Z)-(4-methylbenzylidene) amino) benzamide (3, 0.356 g 1.00 mmol), tartaric acid(0.075 g,0.50 mmol), copper (II) chloride dehydrate (0.170 g, 1.00 mmol) in aqueous methanol (20 ml) as deep red coloured solid (0.83 g) in 78.4% yield. IR (Nujol): 3743 cm^{-1} (N-H), 3278 cm^{-1} (O-H), 3262 cm^{-1} (C-H), 1658 cm^{-1} (C=O), 1625 cm^{-1} (C=N), 1153 cm^{-1} (C-O); MS (relative abundance) m/z = 1059.0 $[M+H]^+$; Anal. % Calc for $C_{50}H_{54}Cu_2N_6O_{12}$: C 56.76, H 5.14, Cu 12.01, N 7.94, O 18.15; Found: C 56.79, H 5.12, Cu 12.03, N 7.93, O 18.13.

Zinc metal complex (5e)

Obtained from(E)-N-(4-methylbenzylidene)-2-((Z)-(4-methylbenzylidene) amino) benzamide (3, 0.356 g 1.00 mmol), tartaric acid(0.075 g,0.50 mmol), anhydrous zinc (II) chloride (0.136 g, 1.00 mmol) in dry methanol (20 ml) as light green coloured solid (0.87 g) in 76.9% yield. IR (Nujol): 3757 cm^{-1} (N-H), 3281 cm^{-1} (O-H), 3263 cm^{-1} (C-H), 1662 cm^{-1} (C=O), 1621 cm^{-1} (C=N), 1135 cm^{-1} (C-O); MS (relative abundance) m/z = 1132.0 $[M+H]^+$;

Anal. % Calc for $C_{50}H_{46}Cl_4Zn_2N_6O_8$: C 53.07, H 4.10, Cl 12.53, N 7.43, O 11.31, Zn 11.56; Found: C 53.11, H 4.13, Cl 12.50, N 7.44, O 11.31, Zn 11.53.

Zinc metal complex (5f)

Obtained from(E)-N-(4-methylbenzylidene)-2-((Z)-(4-methylbenzylidene) amino) benzamide (3, 0.356 g 1.00 mmol), tartaric acid(0.075 g,0.50 mmol), anhydrous zinc (II) chloride (0.136 g, 1.00 mmol) in aqueous methanol (20 ml) as light green coloured solid (0.81 g) in 76.3% yield. IR (Nujol): 3747 cm^{-1} (N-H), 3289 cm^{-1} (O-H), 3258 cm^{-1} (C-H), 1668 cm^{-1} (C=O), 1619 cm^{-1} (C=N), 1160 cm^{-1} (C-O); MS (relative abundance) m/z = 1062.0 $[M+H]^+$; Anal. % Calc for $C_{50}H_{54}Zn_2N_6O_{12}$: C 56.56, H 5.13, N 7.92, O 18.08, Zn 12.32; Found: C 56.49, H 5.15, N 7.93, O 18.10, Zn 12.33.

Nickel metal complex (5g)

Obtained from(E)-N-(4-methylbenzylidene)-2-((Z)-(4-methylbenzylidene) amino) benzamide (3, 0.356 g 1.00 mmol), tartaric acid(0.075 g,0.50 mmol), nickel (II) chloride hexahydrate (0.237 g, 1.00 mmol) in dry methanol (20 ml) as light purple coloured solid (0.79 g) in 70.6% yield. IR (Nujol): 3737 cm^{-1} (N-H), 3285 cm^{-1} (O-H), 3257 cm^{-1} (C-H), 1658 cm^{-1} (C=O), 1621 cm^{-1} (C=N), 1126 cm^{-1} (C-O); MS (relative abundance) m/z = 1119.0 $[M+H]^+$; Anal. % Calc for $C_{50}H_{46}Cl_4N_6Ni_2O_8$: C 53.71. H 4.15, Cl 12.68, N 7.52, Ni 10.50, O 11.45; Found: C 53.68. H 4.17, Cl 12.71, N 7.50, Ni 10.47, O 11.48.

Nickel metal complex (5h)

Obtained from(E)-N-(4-methylbenzylidene)-2-((Z)-(4-methylbenzylidene) amino) benzamide (3, 0.356 g 1.00 mmol), tartaric acid(0.075 g,0.50 mmol), nickel (II) chloride hexahydrate (0.237 g, 1.00 mmol) in aqueous methanol (20 ml) as dark purple coloured solid (0.76 g) in 72.4% yield. IR (Nujol): 3744 cm^{-1} (N-H), 3281 cm^{-1} (O-H), 3249 cm^{-1} (C-H), 1668 cm^{-1} (C=O), 1619 cm^{-1} (C=N), 1149 cm^{-1} (C-O); MS (relative abundance) m/z = 1049.0 $[M+H]^+$; Anal. % Calc for $C_{50}H_{54}N_6Ni_2O_{12}$: C 57.28, H 5.19, N 8.02, Ni 11.20, O 18.31; Found: C 57.32, H 5.17, N 8.07, Ni 11.18, O 18.26.

2.2. Biology.

2.2.1. DPPH free radical-scavenging ability assay.

The radical scavenging activities of various synthesized ligand (**3**) and transition metal complexes (**5a-h**) were determined spectrophotometrically by using the DPPH radical as a reagent (100µL) in ethanol, according to the methods followed by Santosh Kumar *et al.*,(different concentration W/V) [26]. The decrease in absorbance which is induced by antioxidants due to the reduction capacity of DPPH radicals, was measured at 517 nm. Series of test solutions were prepared (10 to 50 µL) in DMF, and the mixture was incubated for 30min at the black room temperature, and then absorbance was measured [100µL of the DPPH solution]. The RadicalScavenging Activity (RSA) of each sample was calculated using the following equation.

$$\% RSA = \frac{Ac-Af}{Ac} \times 100$$

where Ac-is the absorbance of the control and Af-is the absorbance of the test sample. The experiment was done in triplicate.

2.2.2. Antimicrobial activity.

All the synthesized transition metal complexes (**5a-h**) along with the intermediate ligand (**3**), were tested for their *in vitro* antibacterial activity against clinical bacterial strains such as *Staphylococcus aureus* (MTCC 96), *Bacillus cereus* (MTCC 8372)(gram-positive), *Klebsiella pneumonia*, *Escherichia coli* (MTCC 724) (gram-negative) by disc diffusion method [27]. First, a suspension solution of the microorganism (0.1ml of 108 cells/ml) was taken in solid media plates provided with filter paper discs. The synthesized compounds were taken in DMF at 50 to 100µg/ml concentration and incubated with above-developed media in an incubator for 24 hours. The inhibition zone was recorded in diameter (mm) using *Tetracycline* as a standard drug. The antifungal activity was carried out through a microdilution method against 4 clinical strains such as *Aspergillus niger* (MTCC 281), *Aspergillus flavus* (MTCC 873), *Fusarium moniliform* (MTCC 156), and *Fusarium oxysporum* (MTCC 284) by using *Nystatin* as a standard drug. The activity was performed using 96-well plates containing a Mueller-Hinton broth medium (MHB) blend and different concentrations of compounds at 28 ^{o}C. Minimum inhibition concentration (MIC) and minimal bacterial concentration (MBC)/minimal fungicidal concentration (MFC) on agar plates were determined after incubation over 24 hours. The MIC and MBC/MFC values were taken as the mean of three replicates.

3. Results and Discussion

3.1. Chemistry.

All the newly synthesized compounds (**3, 5a-h**) were characterized by IR, NMR, and mass spectral analysis. The structures assigned to the synthesized compounds were also supported by elemental analysis. In IR spectra, appearance of stretching frequency peaks at 3737-3757 cm^{-1} for (N-H), 3249-3269 $cm^{-1}$for (C-H), 3270-3289 $cm^{-1}$for (O-H), 1650-1668 cm^{-1} for (C=O) and 1126-1160 $cm^{-1}$for (C-O) indicated the formation of metal complexes. The appearance of sharp singlets at δ 2.34 ppm for the –CH_3 group and δ8.03 ppm for the –NH- group indicated the formation of **5a**. Two singlets also supported it at δ 8.18 ppm and δ 8.78 ppm for azomethine groups. The appearance of a sharp carbon peak at 20.8 for two methyl groups also supported the formation of complex **5a**. On the other hand, the appearance of a very week signal at 2.17 for the H_2O and –NH- group, in addition to the above characteristic signals, confirmed the formation of hydrated metal complex **5b**.

In mass spectra, all the synthesized metal complexes showed molecular ion peaks (M+1) concerning molecular weight, which supported the structures of the obtained complexes. Similarly, IR, NMR, mass, and elemental analysis supported all the transition metal complexes.

3.2. Biology.

All the newly synthesized ligand (**3**) and transition metal (II) complexes (**5a-h**) were screened for their antioxidant activity and the results obtained are depicted inTable 1. The bioassay of the synthesized compounds envisioned that most of the compounds showed

excellent antioxidant activity. The maximum antioxidant activity was observed in the order of **5e**, **5c**, **5g**, and **5a**. This may be due to the holding of X-type ligands such as chloride by metal (II) complexes. Among the tested compounds, transition metal complexes **5e** and **5c** exhibited excellent interaction with stable DPPH radicals. This may be due to the presence of enzymatic Zn(II) and Cu(II) metal ions in addition to X-type ligands, such as chloride in the metal complex. Compared to other metal complexes having neutral ligand (H_2O) such as **5b**, **5d**, **5f**, **5h,** metal complex **5g** and **5a** holding X type ligand (chloride) exhibited moderate activity, comparable with the standard drug.

All other metal complexes **5b**, **5d**, **5f**, **5h**, including newly synthesized ligand **3**, showed poor antioxidant activity compared to standard drug ascorbic acid. This may be due to the presence of neutral water molecules in the metal complex.

In addition, all the newly synthesized ligand (**3**) and transition metal complexes (**5a-h**) were tested for their antimicrobial efficacy disc diffusion method. The results of antibacterial and antifungal activity are depicted in Table 2 and Table 3, respectively. The measured minimal inhibitory concentration (MIC), minimal bactericidal concentration (MBC), and minimal fungicidal concentration (MFC) values are represented in Table 4. The antimicrobial assay carried out against microbes was comparable with antioxidant activity. Among all five tested bacterial strains, gram-positive and gram-negative bacteria were inhibited mostly by metal complexes **5e**, **5c**, **5g,** and **5a**. The results envisioned that the complexes**5e** and **5c** showed excellently and **5g** and **5a** showed good antimicrobial activity against all the tested strains of microbes. The activity is mainly affected by enzymatic metals such as Zn(II), Cu(II), and X-type ligand in the final compounds. Including the newly synthesized ligand (**3**), all other metal complexes with neutral ligands (water) such as 5b, 5d, 5f, and 5h showed moderate antimicrobial activity compared to standard drugs.

Table 1. Antioxidant activity of synthesized ligand (3) and transition metal (II) complexes (5a-h).

Products	**M**	**X**	**% DPPH radical scavenging assay**				
			10 µg/mL ± SD	**20** µg/mL ± SD	**30** µg/mL ± SD	**40** µg/mL ± SD	**50** µg/mL ± SD
3	-	-	19±0.032	21±0.012	27 ± 0.018	31 ± 0.097	42±0.037
5a	Co	Cl	25±0.017	28±0.021	35 ± 0.002	43 ± 0.082	46±0.029
5b	Co	H_2O	18±0.024	19±0.009	31 ± 0.027	39 ± 0.275	44±0.121
5c	Cu	Cl	30±0.025	33±0.033	39 ± 0.068	50 ± 0.006	55±0.033
5d	Cu	H_2O	22±0.019	24±0.025	29 ± 0.009	40 ± 0.053	43±0.013
5e	Zn	Cl	33±0.027	39±0.017	43 ± 0.023	54 ± 0.112	64±0.039
5f	Zn	H_2O	22±0.115	25±0.032	33 ± 0.039	39 ± 0.087	42±0.012
5g	Ni	Cl	27±0.028	30±0.008	37 ± 0.076	46 ± 0.053	52±0.023
5h	Ni	H_2O	15±0.141	18±0.123	28 ± 0.032	41 ± 0.042	40±0.043
Ascorbic acid	--------		31 ± 0.033	36 ± 0.021	43 ± 0.026	51 ± 0.033	58 ± 0.005

Values are expressed as mean ± standard deviation (n=3).

Table 2. Inhibitory zone (diameter) mm of synthesized ligand (3) and transition metal (II) complexes (5a-h) against tested bacterial strains.

Products	M	X	Antibacterial activity							
			Gram-positive				Gram-negative			
			B. cereus		*S. aureus*		*E. coli*		*K. pneumonia*	
			50 µg/mL ± SD	**100** µg/mL ± SD	**50** µg/mL ± SD	**100** µg/mL ± SD	**50** µg/mL ± SD	**100** µg/mL ± SD	**50** µg/mL ± SD	**100** µg/mL ± SD
3	-	-	04 ± 0.23	09 ± 0.17	06 ± 0.15	10 ± 0.18	12± 0.09	15 ± 0.12	08 ± 0.12	14 ± 0.11
5a	Co	Cl	10 ± 0.25	11 ± 0.25	14 ± 0.24	15 ± 0.24	13 ± 0.22	09 ± 0.26	04± 0.08	06 ± 0.24
5b	Co	H_2O	05 ± 0.09	06 ± 0.15	04 ± 0.14	05 ± 0.17	07 ± 0.17	09 ± 0.08	02± 0.19	04 ± 0.13
5c	Cu	Cl	14 ± 0.16	13 ± 0.11	15 ± 0.21	19 ± 0.19	21 ± 0.07	17 ± 0.06	15± 0.12	18 ± 0.15
5d	Cu	H_2O	04 ± 0.12	07 ± 0.26	05 ± 0.32	09 ± 0.22	07 ± 0.14	07 ± 0.16	02 ± 0.10	07 ± 0.19
5e	Zn	Cl	16 ± 0.07	19 ± 0.21	17 ± 0.09	19 ± 0.14	23 ± 0.08	29 ± 0.18	17± 0.19	21 ± 0.06
5f	Zn	H_2O	03 ± 0.14	05 ± 0.24	06 ± 0.25	08 ± 0.15	07 ± 0.14	10 ± 0.09	07 ± 0.24	09 ± 0.27
5g	Ni	Cl	12 ± 0.17	21 ± 0.17	09 ± 0.25	18 ± 0.12	18 ± 0.23	19 ± 0.14	07 ± 0.32	14 ± 0.06
5h	Ni	H_2O	06 ± 0.09	10 ± 0.32	08 ± 0.15	12 ± 0.19	10 ± 0.24	13 ± 0.28	04 ± 0.16	06 ± 0.29
Tetracycline	--------		12 ± 0.19	20± 0.07	10 ± 0.21	19 ± 0.28	19 ± 0.08	28 ± 0.09	13 ± 0.22	21 ± 0.24

[a]Zone of inhibition (Mean six replicate ± standard deviation).

Table 3. Inhibitory zone (diameter) mm of synthesized ligand (3) and transition metal (II) complexes (5a-h) against tested fungal strains.

Products	Antifungal activity							
	A. flavus		*A. niger*		*F oxysporum*		*F monaliforme*	
	50 µg/mL ± SD	**100** µg/mL ± SD	**50** µg/mL ± SD	**100** µg/mL ± SD	**50** µg/mL ± SD	**100** µg/mL ± SD	**50** µg/mL ± SD	**100** µg/mL ± SD
3	03 ± 0.16	05 ± 0.12	03 ± 0.24	05 ± 0.18	05 ± 0.14	05 ± 0.16	06 ± 0.27	07 ± 0.26
5a	05 ± 0.11	06 ± 0.21	05 ± 0.16	06 ± 0.13	05 ± 0.18	08 ± 0.16	06± 0.28	09 ± 0.09
5b	10 ± 0.11	12 ± 0.21	08 ± 0.16	14 ± 0.13	08 ± 0.18	10 ± 0.16	06± 0.28	09 ± 0.09
5c	08 ± 0.13	09 ± 0.12	04 ± 0.13	05 ± 0.26	05 ± 0.33	07 ± 0.22	03± 0.14	06 ± 0.17
5d	02 ± 0.19	03 ± 0.15	03 ± 0.29	05 ± 0.13	04 ± 0.11	03 ± 0.29	04± 0.13	04 ± 0.19
5e	11 ± 0.09	14 ± 0.26	09 ± 0.07	15 ± 0.17	14 ± 0.11	19 ± 0.18	08± 0.07	12 ± 0.22
5f	03 ± 0.22	05 ± 0.07	02 ± 0.23	06 ± 0.15	02 ± 0.17	03 ± 0.38	04 ± 0.27	06 ± 0.22
5g	06± 0.22	09 ± 0.21	07 ± 0.08	10± 0.12	08 ± 0.14	11 ± 0.22	06± 0.25	07 ± 0.32
5h	04 ± 0.06	05 ± 0.34	04 ± 0.11	07 ± 0.22	03 ± 0.22	05 ± 0.23	02 ± 0.25	03 ± 0.32
Nystatin	15 ± 0.12	20 ± 0.23	12 ± 0.09	27 ± 0.21	15 ± 0.18	32 ± 0.16	11± 0.07	26 ± 0.14

[a] Zone of inhibition (Mean six replicate ± standard deviation).

Table 4. The minimal inhibitory concentration (MIC), minimal bactericidal concentration (MBC), and minimal fungicidal concentration (MFC) in µg/mL of synthesized ligand (3) and transition metal (II) complexes (5a-h) against tested strains.

Compounds	Antibacterial activity								Antifungal activity							
	Gram-positive				Gram-negative											
	B. cereus		*S. aureus*		*E. coli*		*K. pneumonia*		*A. flavus*		*A. niger*		*F. oxysporum*		*F. monaliforme*	
	MIC	**MBC**	**MIC**	**MBC**	**MIC**	**MBC**	**MIC**	**MBC**	**MIC**	**MFC**	**MIC**	**MFC**	**MIC**	**MFC**	**MIC**	**MFC**
3	40	255	50	270	35	195	45	240	35	215	50	265	35	195	30	245
5a	35	190	35	205	35	175	30	170	45	225	35	230	35	230	30	210
5b	50	170	55	255	45	220	40	255	40	245	35	225	35	175	40	240
5c	25	145	30	165	20	125	25	145	30	130	25	190	20	155	25	135
5d	55	240	45	230	50	270	55	260	30	225	55	235	50	250	40	270
5e	20	135	25	135	25	160	20	125	20	125	20	120	20	140	25	120
5f	55	210	40	220	45	250	40	230	45	230	40	210	40	210	35	220
5g	30	160	30	165	20	205	25	165	20	185	25	150	25	155	20	130
5h	40	230	35	260	50	220	40	260	40	235	50	245	45	220	45	230

Compounds	Antibacterial activity								Antifungal activity							
	Gram-positive				Gram-negative											
	B. cereus		*S. aureus*		*E. coli*		*K. pneumonia*		*A. flavus*		*A. niger*		*F. oxysporum*		*F. monaliforme*	
	MIC	MBC	MIC	MBC	MIC	MBC	MIC	MBC	MIC	MFC	MIC	MFC	MIC	MFC	MIC	MFC
Tetracycline	5	135	15	110	15	130	9	110	---	---	---	---	---	---	---	---
Nystatin	---	---	---	---	---	---	---	---	07	120	12	120	20	110	10	110

[a](Mean six replicate ± standard deviation).

4. Conclusions

We have described the simple synthetic method for the preparation of transition metal (II) complexes by treating series of transition metals such as Co(II), Cu(II), Zn(II), Ni(II) with a simple Schiff's base which was initially obtained by the condensation reaction of 4-methylbenzaldehyde with 2-aminobenzohydrazide at elevated temperature. The newly synthesized ligand has been screened for antioxidant, antimicrobial, and antifungal activities along with its metal complexes. The bioassay of the synthesized compounds envisioned that **5e** and **5c** emerged as excellent antimicrobial agents.

Funding

This research received no external funding.

Acknowledgments

The authors are thankful for the V V Sangh'S Ballari and Vijayanagar College Hosapete for providing necessary facilities and constant encouragement.

Conflicts of Interest

The authors declare no conflict of interest.

References

1. Matsumoto, Y.; Sawamura, J.; Murata, Y.; Nishikata, T.; Yazaki, R.; Ohshima, T. Amino Acid Schiff Base Bearing Benzophenone Imine As a Platform for Highly Congested Unnatural α-Amino Acid Synthesis. *J. Am. Chem. Soc*.**2020,** *142*, 8498-8505, https://doi.org/10.1021/jacs.0c02707.
2. MacLachlan, M.J.; Park, M.K.; Thompson, L.K. Coordination compounds of Schiff-base ligands derived from diaminomaleonitrile (DMN): mononuclear, dinuclear, and macrocyclic derivatives. *Inorg. Chem.* **1996,** *35*, 5492-5499, https://doi.org/10.1021/ic960237p.
3. Hamil, A.; Khalifa, K.M.; Almutaleb, A.A.; Nouradean, M.Q. Synthesis, Characterization, and antibacterial activity studies of some transition metal chelates of Mn (II), Ni (II), and Cu (II) with Schiff base derived from diacetylmonoxime with O-phenylenediamine. *Adv. J. Chem. A.* **2020,** *3*, 524-533, https://dx.doi.org/10.33945/SAMI/AJCA.2020.4.13.
4. Schiff, H. MittheilungenausdemUniversitätslaboratorium in Pisa: eineneueReiheorganischer Basen. *Justus Liebigs Ann. Chem.* **1864**, *131*, 118-119, https://doi.org/10.1002/jlac.18641310113.
5. Shamsipur, M.; Ghiasvand, A.R.; Sharghi, H.; Naeimi. H. Solid-phase extraction of ultra-trace copper (II) using octadecyl silica membrane disks modified by a naphthol-derivative Schiff's base. *Anal. Chim. Acta.* **2000**, *408*, 271-277, https://doi.org/10.1016/S0003-2670(99)00873-9.
6. Mahmoudi, F.; Farhadi, S.; Dusek, M.; Poupon, M. Synthesis, Spectroscopy and X-ray Crystallography Structure of Pyridine 4-Carbaldehyde Semicarbazone Schiff Base Ligand. *Adv. J. Chem. A.***2020,** *3*, 534-541, https://doi.org/10.33945/SAMI/AJCA.2020.4.14.

7. Xia, Y.; Wang, J. Transition-metal-catalyzed cross-coupling with ketones or aldehydes via N-tosylhydrazones. *J. Am. Chem. Soc.***2020,** *142*, 10592-10605, https://doi.org/10.1021/jacs.0c04445.
8. Hamed, A.A.; Abdelhamid, I.A.; Saad, G.R.; Elkady, N.A.; Elsabee, M.Z. Synthesis, characterization, and antimicrobial activity of a novel chitosan Schiff bases based on heterocyclic moieties. *Int. J. Biol. Macromol.* **2020,** *153*, 492-501, https://doi.org/10.1016/j.ijbiomac.2020.02.302.
9. Magalhaes, T.F.F.; da Silva, C.M.; Dos Santos, L.B.; Santos, D.A.; Silva, L.; Fuchs, B.B.; Mylonakis, E.; Martins, C.V.; de Resende-Stoianoff, M.A.; de Fatima, A. Cinnamyl Schiff bases: synthesis, cytotoxic effects and antifungal activity of clinical interest. *Lett. Appl. Microbiol.* **2020,** *71*, 490-497, https://doi.org/10.1111/lam.13356.
10. Hassan, A.M.; Osman Said, A.; Heakal, B.H.; Younis, A.; Abdelmoaz, M.A.; Abdrabou, M.M. Conventional and Microwave-Assisted Synthesis, Antimicrobial and Antitumor Studies of Tridentate Schiff Base Derived from O-vanillin and Phenyl Urea and its Complexes. *Adv. J. Chem. A.* **2020**, *3*, 621-638, https://doi.org/10.22034/AJCA.2020.105996.
11. Zhu, S.; Xu, S.; Yi, X.; Wang, J.; Zhao, Z.; Jiang, J. High value-added application of turpentine as a potential renewable source for the synthesis of heterocyclic Schiff base derivatives of cis-1, 8-p-menthane-diamine serving as botanical herbicides. *Ind. Crops. Prod.* **2018**, *115*, 111-116, https://doi.org/10.1016/j.indcrop.2018.02.021.
12. Beyazit, N.; Çakran, H.S.; Cabir, A.; Akiscan, Y.; Demetgul, C. Synthesis, characterization and antioxidant activity of chitosan Schiff base derivatives bearing (−)-gossypol. *Carbohydr. Polym.* **2020,** *240*, 116333, https://doi.org/10.1016/j.carbpol.2020.116333.
13. Yuksel, E.; Bilici, A.; Gecibesler, I.H.; Kaya. I. Synthesis and antioxidant activities of phenolic Schiff base monomers and polymers. *Can. J. Chem.* **2020,** *98*, 151-157, https://doi.org/10.1139/cjc-2019-0307.
14. Tokali, F.S.; Taslimi, P.; Usanmaz, H.; Karaman, M.; Sendil, K. Synthesis, characterization, biological activity and molecular docking studies of novel Schiff bases derived from thiosemicarbazide: Biochemical and computational approach. *J. Mol. Struct.* **2021**, *1231*, 129666, https://doi.org/10.1016/j.molstruc.2020.129666.
15. Joshi, R.; Kumari, A.; Singh, K.; Mishra, H.; Pokharia, S. Triorganotin (IV) complexes of Schiff base derived from 1, 2, 4-triazole moiety: Synthesis, spectroscopic investigation, DFT studies, antifungal activity, and molecular docking studies. *J. Mol. Struct*. **2020,** *1206*, 127639, https://doi.org/10.1016/j.molstruc.2019.127639.
16. Frei, A.; King, A.P.; Lowe, G.J.; Cain, A.K.; Short, F.L.; Dinh, H.; Elliott, A.G.; Zuegg, J.; Wilson, J.J.; Blaskovich, M.A. Nontoxic Cobalt (III) Schiff Base Complexes with Broad-Spectrum Antifungal Activity. *Chem. Eur. J.* **2021,** *27*, 2021-2029, https://dx.doi.org/10.1002%2Fchem.202003545.
17. Singh, K.; Barwa, M.S.; Tyagi, P. Synthesis and characterization of cobalt (II), nickel (II), copper (II), and zinc (II) complexes with Schiff base derived from 4-amino-3-mercapto-6-methyl-5-oxo-1, 2, 4-triazine. *Eur. J. Med. Chem.* **2007**, *42*, 394-402, https://doi.org/10.1016/j.ejmech.2006.10.016.
18. Barbosa, H.F.; Attjioui, M.; Ferreira, A.P.; Moerschbacher, B.M.; Cavalheiro, E.T. New series of metal complexes by amphiphilic biopolymeric Schiff bases from modified chitosans: Preparation, characterization, and effect of molecular weight on its biological applications. *Int. J. Biol. Macromol.* **2020,** *145*, 417-428, https://doi.org/10.1016/j.ijbiomac.2019.12.153.
19. Buldurun, K.; Turan, N.; Bursal, E.; Mantarci, A.; Turkan, F.; Taslimi, P.; Gulcin, I. Synthesis, spectroscopic properties, crystal structures, antioxidant activities and enzyme inhibition determination of Co (II) and Fe (II) complexes of Schiff base. *Res. Chem. Intermed*. **2020,** *46*, 283-297, https://doi.org/10.1007/s11164-019-03949-3.
20. Buldurun, K.; Turan, N.; Bursal, E.; Aras, A.; Mantarci, A.; Colak, N.; Turkan, F.; Gulcin, I. Synthesis, characterization, powder X-ray diffraction analysis, thermal stability, antioxidant properties and enzyme inhibitions of M (II)-Schiff base ligand complexes. *J. Biomol. Struct. Dyn.* **2020,** *4*, 1-8, https://doi.org/10.1080/07391102.2020.1802340.
21. Savir, S.; Wei, Z.J.; Liew, J.W.; Vythilingam, I.; Lim, Y.A.; Saad, H.M.; Sim, K.S.; Tan, K.W. Synthesis, cytotoxicity and antimalarial activities of thiosemicarbazones and their nickel (II) complexes. *J. Mol. Struct*. **2020,** *1211*, 128090, https://doi.org/10.1016/j.molstruc.2020.128090.
22. Malekshah, R.E.; Shakeri, F.; Khaleghian, A.; Salehi, M. Developing a biopolymeric chitosan supported Schiff-base and Cu (II), Ni (II) and Zn (II) complexes and biological evaluation as pro-drug. *Int. J. Biol. Macromol.* **2020,** *152*, 846-861, https://doi.org/10.1016/j.ijbiomac.2020.02.245.

23. Szklarzewicz, J.; Jurowska, A.; Matoga, D.; Kruczała, K.; Kazek, G.; Mordyl, B.; Sapa, J.; Papiez, M. Synthesis, coordination properties and biological activity of vanadium complexes with hydrazone Schiff base ligands. *Polyhedron*. **2020,** *185*, 114589, https://doi.org/10.1016/j.poly.2020.114589.
24. Rambabu, A.; Ganji, N.; Daravath, S.; Venkateswarlu, K.; Rangan, K. Mononuclear Co (II), Ni (II) and Cu (II) complexes of the Schiff base, 2-(((4-trifluoromethoxy) phenylimino) methyl)-6-tert-butylphenol: Synthesis, spectroscopic characterization, X-ray study, and biological evaluation. *J. Mol. Struct*. **2020,** *1199*, 127006, https://doi.org/10.1016/j.molstruc.2019.127006.
25. Bian, M.; Wang, X.; Sun, Y.; Liu, W. Synthesis and biological evaluation of gold (III) Schiff base complexes for the treatment of hepatocellular carcinoma through attenuating TrxR activity. *Eur. J. Med. Chem.***2020,** *193*, 112234, https://doi.org/10.1016/j.ejmech.2020.112234.
26. Santosh Kumar, S.; Priyadarsini, K.I.; Sainis, K.B. Free radical scavenging activity of vanillin and o-vanillin using 1,1-diphenyl-2-picrylhydrazyl (DPPH) radical. *Redox. Rep.* **2002**, *7*, 35–40, https://doi.org/10.1179/135100002125000163.
27. Andrews, J.M. BSAC standardized disc susceptibility testing method (version 7). *J. Antimicrob. Chemother.* **2008**, *62*, 256-278, https://doi.org/10.1093/jac/dkn194.