Scientific paper

Synthesis, X-ray Crystal Structures and Antibacterial Activity of Copper(II) Complexes Derived from Halo Substituted Hydrazones

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Abstract

Schiff bases bearing halo substituent groups have interesting biological activities. In this work, two new hydrazone type Schiff bases *N*'-(3,5-dibromo-2-hydroxybenzylidene)-3-methylbenzohydrazide (HL^1) and *N*'-(4-chloro-2-hydroxybenzylidene)-4-fluorobenzohydrazide (HL^2), bearing bromo, chloro and fluoro substituent groups were synthesized. The compounds reacts with various copper salts to form three copper(II) complexes with interesting structures. The complexes are $[CuBrL^1]\cdot H_2O$ (**1**·H_2O), $[CuClL^2]\cdot CH_3OH$ (**2**·CH_3OH) and $[Cu_2Br_2L^2_2(CH_3OH)_2]$ (**3**). The hydrazones and three copper complexes were characterized by physico-chemical methods such as elemental analysis, infrared and electronic spectroscopy. The free hydrazones were also characterized by ^1H NMR spectroscopy, and structures of H_2L^1 and the complexes were further confirmed by single crystal X-ray determination. The hydrazone ligands in the complexes coordinate to Cu ions through phenolate oxygen, imino nitrogen and carbonyl oxygen atoms. The Cu ion in complex **1** is in square planar coordination, in complex **2** is in square pyramidal coordination, and in complex **3** is in octahedral coordination. The complexes show good antibacterial activities on the bacterial strains *Bacillus subtilis*, *Staphylococcus aureus* and *Escherichia coli*, while weak activity on the fungal strain *Candida albicans* and no activity on *Aspergillus niger*.

Keywords: Schiff base; Copper complexes; X-ray crystal structure; Antimicrobial activity.

1. Introduction

Schiff bases and their metal complexes have displayed significant biological activities like antibacterial, antifungal, anticancer, antitubercular, anti-inflammatory, antitumor, and antioxidant, *etc*.[1] Hydrazone compounds bearing –CH=N–NH–C(O)–functional group are a special kind of Schiff bases. These compounds show interesting biological activities, especially in the fields of antibacterial and antifungal action.[2] Hydrazone compounds readily coordinate to metal ions and form complexes with various structures and biological properties.[3] Copper is a biological dependence trace element in human beings, animals and most plants. Copper may contribute to beneficial effects as part of the standard therapeutic management of chronic obstructive pulmonary disease.[4] Copper is routinely supplemented to weanling pig diets at concentrations above nutritional requirements to enhance growth performance.[5] In addition, copper plays an important role in the pathophysiology of fibromyalgia.[6] Copper complexes with Schiff bases have shown interesting antibacterial activities.[7] Notably, compounds bearing halo substituent groups have been reported to possess enhanced biological activities.[8] Our research group has continuously been interested in biological activities of Schiff base complexes and has reported the synthesis and antimicrobial activities of complexes with single halo-substituted Schiff bases.[9] In addition, some hydrazones and their complexes with metals often have diverse biological and pharmaceutical activities. However, up to now the biological activity and interactions of metal complexes with hydrazone ligands bearing both chloro and fluoro substituent groups have seldom been reported. This aroused our interest in the synthesis of hydrazone and its copper(II) complexes. In this work, two Schiff bases *N*'-(3,5-dibromo-2-hydroxybenzylidene)-3-methylbenzohydrazide (HL^1) and *N*'-(4-chloro-2-hydroxyben-

zylidene)-4-fluorobenzohydrazide (HL^2), were used to prepare copper complexes $[CuBrL^1]\cdot H_2O$ (**1**), $[CuClL^2]\cdot CH_3OH$ (**2**) and $[Cu_2Br_2L^2{}_2(CH_3OH)_2]$ (**3**). Antibacterial and antifungal activities of the compounds were determined.

2. Experimental Section

2. 1. Materials and Measurements

3,5-Dibromosalicylaldehyde, 4-chlorosalicylaldehyde, 3-methylbenzohydrazide and 4-fluorobenzohydrazide with AR grade were purchased from Macklin Chemical Co. Ltd. Copper bromide, copper chloride and methanol with AR grade were obtained from Liaodong Chemical Co. Ltd. Kanamycin, penicillin G and ketokonazole were obtained from Aladdin Chemical Co. Ltd. All solvents and other chemicals used were commercially available and used as received. CHN elemental analyses were performed on a Perkin-Elmer 240C elemental analyzer. IR spectra were recorded on a Nicolet AVATAR 360 spectrophotometer as KBr pellets in the 4000–400 cm^{-1} region. UV-Vis spectra were recorded on a Lambda 35 spectrophotometer. DDS-11A conductivity meter was used to determine molar conductivity values of the complexes. Bruker 300 MHz instrument was used to determine ^1H NMR of the free Schiff bases. Single crystal structures of the three copper complexes were determined with a Bruker Apex II CCD diffractometer.

2. 2. Synthesis of the Schiff Bases

2. 2. 1. HL^1

3,5-Dibromosalicylaldehyde (2.79 g, 0.010 mol) and 3-methylbenzohydrazide (1.50 g, 0.010 mol) were dissolved in 50 mL methanol. The reaction mixture was refluxed for 10 min, and with the solvent removed by distillation under reduced pressure. The white powder was re-crystallized from methanol to give crystalline product of HL^1. Yield: 3.85 g (94%). $C_{15}H_{12}Br_2N_2O_2$: calcd. for C 43.72; H 2.94; N 6.80%; found: C 43.56; H 3.02; N 6.73%. Characteristic IR: $\bar{\upsilon}$ = 3215 ν(NH), 1655 ν(C=O), 1612 ν(C=N) cm^{-1}. UV-Vis data (λ_{max}, ε): 220 nm, 7,130 L mol^{-1} cm^{-1}; 292 nm, 19,870 L mol^{-1} cm^{-1}; 305 nm, 19,125 L mol^{-1} cm^{-1}; 336 nm, 8,150 L mol^{-1} cm^{-1}; 403 nm, 3,457 L mol^{-1} cm^{-1}. ^1H NMR (300 MHz, d_6-DMSO): 12.69 (s, 1H, OH), 12.51 (s, 1H, NH), 8.53 (s, 1H, CH=N), 7.83 (d, 1H, ArH), 7.77 (d, 2H, ArH), 7.74 (s, 1H, ArH), 7.47 (s, 1H, ArH), 7.45 (s, 1H, ArH), 2.41 (s, 3H, CH_3) ppm. Single crystals of the compound were obtained by slow evaporation of its methanolic solution.

2. 2. 2. HL^2

4-Chlorosalicylaldehyde (1.56 g, 0.010 mol) and 4-fluorobenzohydrazide (1.54 g, 0.010 mol) were dissolved in 50 mL methanol. The reaction mixture was refluxed for 10 min, and with the solvent removed by distillation under reduced pressure. The white powder was re-crystallized from methanol to give crystalline product of H_2L. Yield: 2.67 g (91%). $C_{14}H_{10}ClFN_2O_2$: calcd. for C 57.45; H 3.44; N 9.57%; found: C 57.26; H 3.51; N 9.65%. Characteristic IR: $\bar{\upsilon}$ = 3276 ν(NH), 1651 ν(C=O), 1619 ν(C=N) cm^{-1}. UV-Vis data (λ_{max}, ε): 236 nm, 16,270 L mol^{-1} cm^{-1}; 275 nm, 9,960 L mol^{-1} cm^{-1}; 286 nm, 14,680 L mol^{-1} cm^{-1}; 300 nm, 16,072 L mol^{-1} cm^{-1}; 330 nm, 16,210 L mol^{-1} cm^{-1}; 372nm, 17,360 L mol^{-1} cm^{-1}. ^1H NMR (300 MHz, d_6-DMSO): 12.13 (s, 1H, OH), 11.53 (s, 1H, NH), 8.63 (s, 1H, CH=N), 8.02 (d, 2H, ArH), 7.61 (d, 1H, ArH), 7.38 (d, 2H, ArH), 6.99 (d, 1H, ArH), 6.97 (s, 1H, ArH) ppm.

2. 3. Synthesis of the Complexes

2. 3. 1. Synthesis of $[CuBrL^1]\cdot H_2O$ ($1\cdot H_2O$)

HL^1 (40mg, 0.10 mmol) was dissolved in 30 mL methanol. Then, copper bromide (22 mg, 0.10 mmol) was added. The reaction mixture was stirred for 30 min at room temperature to afford a clear brown solution. The solution was allowed to slowly evaporate at ambient condition for 7 days to form single crystals, which were collected by filtration and dried in air. Yield: 0.36 g (63%). $C_{15}H_{13}Br_3CuN_2O_3$: calcd. C 31.47; H 2.29; N 4.89%; found: C 31.31; H 2.35; N 4.96%. Characteristic IR: 3156 ν(N-H), 1608 ν(C=N) cm^{-1}. UV-Vis data (λ_{max}, ε): 235 nm, 17,270 L mol^{-1} cm^{-1}; 258 nm, 15,730 L mol^{-1} cm^{-1}; 320 nm, 12,106 L mol^{-1} cm^{-1}; 410 nm, 12,450 L mol^{-1} cm^{-1}.

2. 3. 2. Synthesis of $[CuClL^2]\cdot CH_3OH$ ($2\cdot CH_3OH$) and $[Cu_2Br_2L^2{}_2(CH_3OH)_2]$ (3)

Complex **2** was synthesized by a similar method as described for **1**, but with HL^1 replaced with HL^2 (29 mg, 0.10 mmol), and with copper bromide replaced with copper chloride dihydrate (17 mg, 0.10 mmol). Complex **3** was synthesized by a similar method as described for **1**, but with HL^1 replaced with HL^2.

Complex **2**: 0.31 g (73%). $C_{15}H_{13}Cl_2CuFN_2O_3$: calcd. C 42.62; H 3.10; N 6.63%, found: C 42.78; H 3.03; N 6.51%. Characteristic IR: 1605 ν(C=N) cm^{-1}. UV-Vis data (λ_{max}, ε): 233nm, 15,132 L mol^{-1} cm^{-1}; 265 nm, 10,920 L mol^{-1} cm^{-1}; 297 nm, 7,470 L mol^{-1} cm^{-1}; 310 nm, 8,936 L mol^{-1} cm^{-1}; 325 nm, 8,872 L mol^{-1} cm^{-1}; 387 nm, 10,400 L mol^{-1} cm^{-1}.

Complex **3**: 0.18 g (38%). $C_{30}H_{26}Br_2Cl_2Cu_2F_2N_4O_6$: calcd. C 38.56; H 2.80; N 6.00%, found: C 38.28; H 2.86; N 5.90%. Characteristic IR: 3116 ν(N-H), 1601 ν(C=N) cm^{-1}. UV-Vis data (λ_{max}, ε): 232 nm, 15,105 L mol^{-1} cm^{-1}; 263 nm, 11,338 L mol^{-1} cm^{-1}; 300 nm, 8,730 L mol^{-1} cm^{-1}; 310 nm, 9,920 L mol^{-1} cm^{-1}; 323 nm, 9,210 L mol^{-1} cm^{-1}; 385 nm, 11,276 L mol^{-1} cm^{-1}.

2. 4. Crystal Structure Determination

Selected crystals of HL^1 and the three complexes were mounted on a Bruker Apex II CCD area diffractome-

Table 1. Crystallographic and experimental data for HL^1 and the complexes

Compound	**HL^1**	**$1{\cdot}H_2O$**	**$2{\cdot}CH_3OH$**	**3**
Formula	$C_{16}H_{16}Br_2N_2O_3$	$C_{15}H_{13}Br_3CuN_2O_3$	$C_{15}H_{13}Cl_2CuFN_2O_3$	$C_{30}H_{26}Br_2Cl_2Cu_2F_2N_4O_6$
Formula Weight	444.13	572.54	422.71	934.35
Crystal system	Triclinic	Monoclinic	Monoclinic	Monoclinic
Space group	*P*–1	*Pn*	$P2_1/c$	$P2_1/n$
a /Å	8.9105(11)	11.3302(3)	7.5676(8)	10.9278(12)
b /Å	9.4305(12)	6.2236(2)	29.0846(15)	11.1728(12)
c /Å	11.3352(13)	12.5942(4)	7.8520(8)	14.0764(13)
α /°	95.5630(10)	90	90	90
β /°	111.5710(10)	90.4350(10)	108.3510(10)	97.1610(10)
γ /°	98.6550(10)	90	90	90
V /Å^3	863.61(18)	888.05(5)	1640.3(3)	1705.2(3)
Z	2	2	4	2
D_c /(gcm^{-3})	1.708	2.141	1.712	1.820
μ /mm^{-1}	4.708	7.994	1.684	3.806
F(000)	440	550	852	924
Reflections/parameters	5056/219	9342/231	7829/222	9040/221
Unique reflections	3207	3324	2606	3175
Observed reflections (I^3 2*s*(*I*))	1617	3196	2179	2624
Restraints	0	6	1	1
Goodness-of-fit on F^2	0.993	1.033	1.161	1.031
R_1, wR_2 [$I \geq 2\sigma(I)$]	0.0534, 0.1122	0.0199, 0.0452	0.0344, 0.0935	0.0289, 0.0686
R_1, wR_2 (all data)	0.1279, 0.1417	0.0215, 0.0458	0.0464, 0.1095	0.0399, 0.0736

ter equipped with Mo Kα radiation (λ = 0.71073 Å). Crystal data were collected at room temperature and corrected for Lorentz polarization effects and for linear decay. Multi-scan absorption correction based on ω-scans was applied. Data refinement was carried out by SMART and reduction was SAINT. Structures of the compounds were solved by direct method using SHELXL and successive difference Fourier synthesis.[10] All non-hydrogen atoms were refined anisotropically. The amino and water H atoms in the compounds were located from difference Fourier maps and refined isotropically, with N···H and O···H distances restrained to 0.90(1) and 0.85(1) Å, respectively. The remaining H atoms were fixed geometrically and refined using riding model. Crystallographic data and structure refinement parameters are listed in Table 1.

2. 5. Antimicrobial Assay

Antibacterial activities were assayed on the bacteria strains *B. subtilis*, *S. aureus*, *E. coli* and *P. fluorescens* using Mueller-Hinton medium. Antifungal activities were assayed on the fungi *C. albicans* and *A. niger* using RPMI-1640 medium. MIC values were determined by a colorimetric method using MTT.[11] A stock solution of the tested material (150 *μ*M) dissolved in the mixture of DMSO and water (v:v = 1:99) was prepared and graded quantities (75 *μ*M, 37.5 *μ*M, 18.8 *μ*M, 9.4 *μ*M, 4.7 *μ*M, 2.3 *μ*M, 1.2 *μ*M, 0.59 *μ*M) were incorporated in the corresponding sterilized liquid medium. A specified quantity of the medium containing the tested materials was poured into micro-titration plates. Suspension of the microorganism was prepared to contain 10^5 cfu mL^{-1} and applied to micro-titration plates with the diluted compounds and incubated at 37 °C for 24 h and 48 h for bacteria and fungi, respectively. 50 *μ*L PBS (phosphate buffered saline 0.01 M, pH = 7.4) was added to each well. Incubation was continued at room temperature for 4–5 h. The content of each well was removed, and 100 *μ*L of isopropanol containing 5% 1 M HCl was added to extract the dye. The MIC values were visually determined on each of the micro-titration plates. Kanamycin and penicillin G were used as standards for antibacterial activities, and ketoconazole was used as a standard for antifungal activity.

3. Results and Discussion

3. 1. Chemistry

The Schiff bases HL^1 and HL^2 were synthesized from condensation reaction of 3,5-dibromosalicylaldehyde with 3-methylbenzohydrazide, and 4-chlorosalicylaldehyde-with 4-fluorobenzohydrazide, respectively, in 1:1 molar ratio in methanol. The synthetic procedure for the complexes is shown in Scheme 1. All complexes were synthesized by reaction of the Schiff bases with copper salts in 1:1 molar ratio in methanol.

The elemental analyses of HL^1 and the three copper complexes agree well with the formulae determined by single crystal X-ray determination. The Schiff bases and the copper complexes are soluble in methanol, ethanol,

acetonitrile, DMF and DMSO, but insoluble in water and ethyl ether. Crystals of the complexes are stable in air at room temperature for at least three months. In solution at concentration of 10^{-3} M, complexes **1** and **2** behave as non-electrolytes as evidenced by molar conductivity values of 27–35 Ω^{-1} cm^2 mol^{-1},[12] and complex **3** behaves as a 1:1 electrolyte as evidenced by molar conductivity value of 83 Ω^{-1} cm^2 mol^{-1}. IR data of HL^1 and the copper complexes agree well with their X-ray structures. The free Schiff bases feature typical bands at 1651–1655 and 1612–1619 cm^{-1}, which are attributed to the stretching vibrations of the carbonyl [ν(C=O)] and azomethine [ν(C=N)] groups.[13] In the IR spectra of the complexes, the stretching vibrations of C=N groups shift to lower frequencies when compared with the free Schiff bases, indicating that the ligands coordinate to copper ions *via* azomethine nitrogen. The weak bands in low wavenumbers 400–600 cm^{-1} can be assigned to ν(Cu–O) and ν(Cu–N) stretching vibrations.

3. 2. Structure Description of HL^1

The molecular structure of HL^1 is shown in Figure 1. There is a methanol molecule of crystallization. The distance of the methylidene bond C7–N1 is 1.263(7) Å, which confirmsit as a typical double bond. The shorter distance of the C8–N2 bond (1.363(8) Å) and the longer distance of the C8–O2 bond (1.216(6) Å) for the –C(O)–NH– unit than usual, suggest the presence of conjugation effect in the molecule. The bond lengths in the compound are within normal values.[14] The dihedral angle between the two benzene rings is 5.4(3)°.

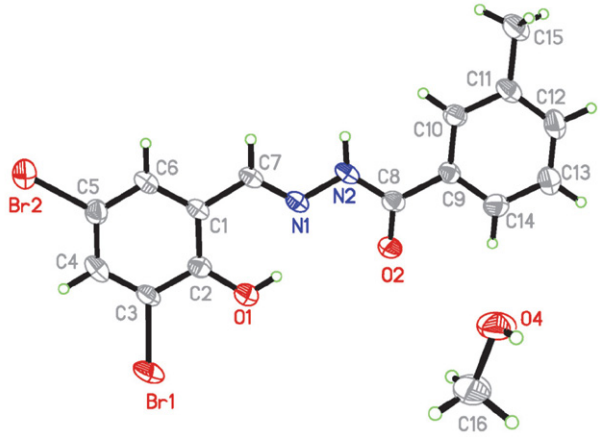
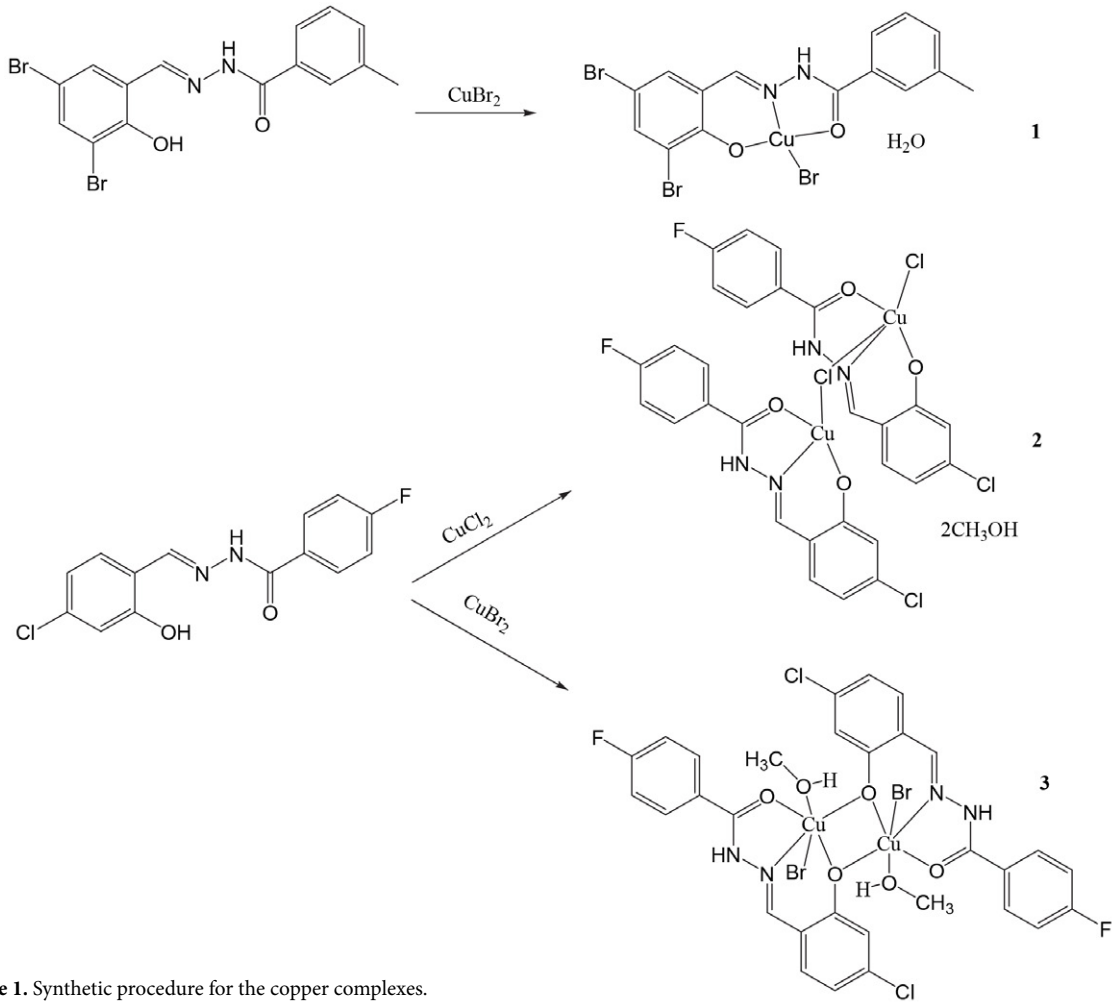


Figure 1. Molecular structure of HL^1. Displacement ellipsoids for non-hydrogen atoms are drawn at the 30% probability level.



Scheme 1. Synthetic procedure for the copper complexes.

3. 3. Structure Description of the Complexes

Selected bond lengths and angles are given in Table 2.

Table 2. Selected bond lengths (Å) and angles (°) for the complexes

1·H_2O			
Cu1–O1	1.883(3)	Cu1–N1	1.950(4)
Cu1–Br3	2.3662(7)	Cu1–O2	1.969(3)
O1–Cu1–N1	91.93(15)	O1–Cu1–O2	171.72(14)
N1–Cu1–O2	80.58(14)	O1–Cu1–Br3	93.80(10)
N1–Cu1–Br3	173.32(11)	O2–Cu1–Br3	93.90(10)
2·CH_3OH			
Cu1–O1	1.893(2)	Cu1–N1	1.944(3)
Cu1–O2	1.987(3)	Cu1–Cl2	2.2539(10)
Cu1–Cl2A	2.810(1)		
O1–Cu1–N1	92.32(11)	O1–Cu1–O2	171.84(11)
N1–Cu1–O2	80.33(11)	O1–Cu1–Cl2	92.27(8)
N1–Cu1–Cl2	162.66(9)	O2–Cu1–Cl2	93.76(8)
O1–Cu1–Cl2A	92.8(1)	O2–Cu1–Cl2A	171.8(1)
N1–Cu1–Cl2A	80.3(1)	Cl2–Cu1–Cl2A	101.1(1)

Symmetry code for A: *x*, 1½ – *y*, –½ + *z*

3			
Cu1–N1	1.934(2)	Cu1–O1	1.9569(18)
Cu1–O1A	1.9678(17)	Cu1–O2	1.9993(19)
Cu1–O3	2.404(2)		
N1–Cu1–O1	91.96(8)	N1–Cu1–O1A	171.43(8)
O1–Cu1–O1A	79.48(8)	N1–Cu1–O2	80.77(8)
O1–Cu1–O2	172.45(8)	O1A–Cu1–O2	107.79(8)
N1–Cu1–O3	93.20(9)	O1–Cu1–O3	92.93(8)
O1A–Cu1–O3	87.64(9)	O2–Cu1–O3	85.49(8)

Symmetry code for A: 1 – *x*, 1 – *y*, 1 – z

3. 2. 1. Complex 1·H_2O

The perspective view of compound **1**·H_2O is shown in Figure 2. The compound is a mononuclear copper complex. There is a water molecule of crystallization.The Cu atom is coordinated by one phenolate oxygen (O1), one carbonyl oxygen (O2) and one imino nitrogen (N1) of a Schiff base ligand, and one bromide ligand (Br1), forming square planar geometry. The Cu–O and Cu–N bond lengths are comparable to the copper complexes derived from Schiff base ligands with square planar coordination.[15] The two benzene rings of the Schiff base ligand form a dihedral angle of 6.9(4)°.

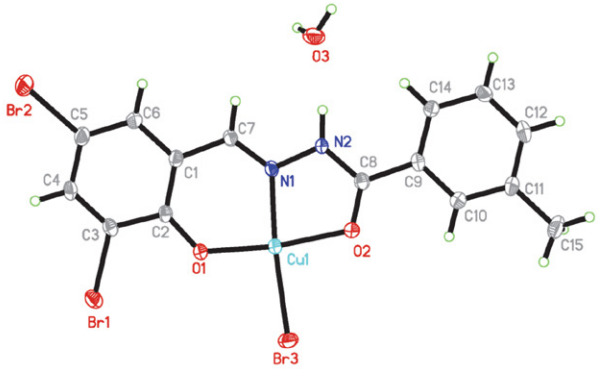


Figure 2. Molecular structure of **1**. Displacement ellipsoids for non-hydrogen atoms are drawn at the 30% probability level.

3. 2. 2. Complex 2·CH_3OH

The perspective view of compound **2**·CH_3OH is shown in Figure3. he Cu atom is coordinated by one oxygen (O1), one carbonyl oxygen (O2) and one imino nitrogen (N1) from a hydrazone ligand, and two chloride ligands (Cl2, Cl2A; symmetry code for A: *x*, 1½ – *y*, –½ + *z*), forming square pyramidal geometry. The Cu–O, Cu–N and Cu–Cl bond lengths are comparable to copper complexes derived from hydrazone ligands with square pyramidal

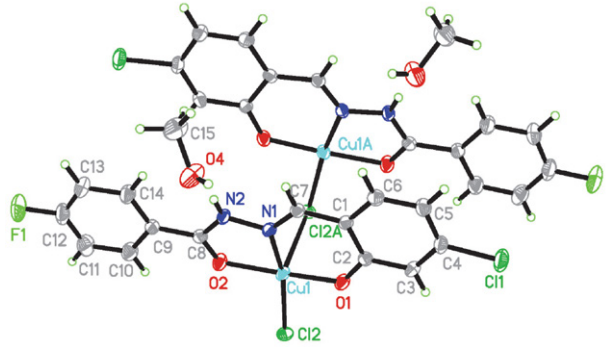


Figure 3. Molecular structure of **2**·CH_3OH. Displacement ellipsoids for non-hydrogen atoms are drawn at the 30% probability level. Symmetry operation: *x*, 1½ – *y*, –½ + *z*.

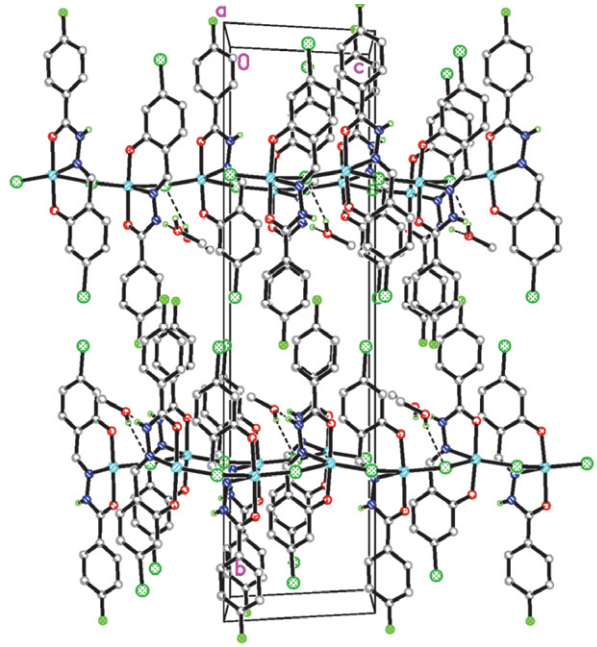


Figure 4. Molecular packing structure of **2**·CH_3OH, viewed along *a* axis. Hydrogen bonds are shown as dashed lines.

coordination.[16] The two benzene rings of hydrazone ligand form a dihedral angle of 6.4(5)°. The [CuL^2] units are linked by chloride ligands to form chains along *c* axis (Figure 4).

3. 2. 3. Complex 3

The perspective view of complex **3** is shown in Figure 5. The compound is a dinuclear copper complex. The complex molecule possesses crystallographic inversion center symmetry. The center is located at the midpoint of two Cu atoms. The metal ions are connected by two phenolate oxygen atoms from two anionic hydrazone ligands, exhibiting two μ_2-O bridges. The distance of two Cu atoms is 3.0179(6) Å. The Cu atom is coordinated by two phenolate oxygens (O1, O1A; symmetry code for A: 1 – *x*, 1 – *y*, 1 – *z*), one carbonyl oxygen (O2) and one imino nitrogen (N1) from two symmetry related hydrazone ligands, one methanol oxygen (O3), and one bromide ligand (Br1), forming octahedral geometry. The Cu–O and Cu–N bond lengths are comparable to the copper complexes derived from hydrazone ligands with octahedral coordination.[17] The two benzene rings of the hydrazone ligand form a dihedral angle of 2.9(3)°.

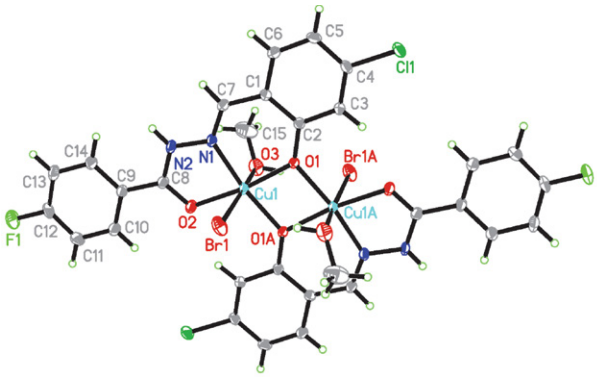


Figure 5. Molecular structure of **3**. Displacement ellipsoids for non-hydrogen atoms are drawn at the 30% probability level. Symmetry operation: 1 – *x*, 1 – *y*, 1 – *z*.

3. 3. Antimicrobial Activity

The hydrazones and three copper complexes were assayed for their *in vitro* antibacterial activity against two Gram(+) bacterial strains *Bacillus subtilis* and *Staphylococcus aureus*, and two Gram(–) bacterial strains *Escherichia coli* and *Pseudomonas fluorescens* by MTT method. The MIC (minimum inhibitory concentration) values are summarized in Table 3. Kanamycin and penicillin G were assayed as references. HL^1 has weak activity against *B. subtilis*, *S. aureus* and *E. coli*, and inactive against the remaining strains. HL^2 has good activity against *B. subtilis*, while weak activity against *S. aureus* and *E. coli*, and inactive against the remaining strains. A comparative study of the ligands and complexes indicates that the copper complexes exhibit higher antibacterial activity than the free ligands. This is caused by greater lipophilic nature of complexes than free ligands. The potent antibacterial activity of complexes could be attributed to the fact that metal complexes have long been known to undergo ligand-substitution reactions with biomolecular targets. Such increased activity of the metal chelates can be explained on the basis of chelating theory.[18] There is a decrease in the polarity of the metal ion significantly after chelation, because of the partial sharing of its positive charge with the donor group and also due to π-electron delocalization on the whole chelate ring. Complexes **1** and **3** have strong activity against *B. subtilis* and *S. aureus*, and weak activity against *E. coli* and *P. fluorescens*. Complex **2** is more active than the other two complexes, which has strong activity against *B. subtilis*, *S. aureus* and *E. coli*, and medium activity against *P. fluorescens*. This indicates that bromide ligand doesn't influence the activities. Instead, the chloride ligand may improve the antibacterial activities. In addition, the chloro and fluoro substitute groups in the ligands of complexes **2** and **3** may contribute to the increasing of antibacterial activities against *B. subtilis* and *S. aureus* than the bromo and methyl substitute groups of complex **1**. Complex **2** has similar activity against *B. subtilis*, *S. aureus* and *E. coli* as the references kanamycin and penicillin G.

Antifungal activities of the compounds were assayed on two fungal strains *Candida albicans* and *Aspergillus niger* by MTT method. Ketoconazole was used as a reference. As a result, complexes **2** and **3** have weak activity against *C. albicans*, while no activity on *A. niger*.

Table 3. The MIC values (μM) of the compounds

Tested material	*B. subtilis*	*S. aureus*	*E. coli*	*P. fluorescens*	*C. albicans*	*A. niger*
1·H_2O	4.7	9.4	18.8	37.5	>150	>150
2·CH_3OH	1.2	2.3	4.7	18.8	75	>150
3	2.3	4.7	18.8	75	75	>150
HL^1	18.8	37.5	37.5	>150	>150	>150
HL^2	4.7	18.8	37.5	>150	>150	>150
Kanamycin	0.6	2.3	4.7	4.7		
Penicillin G	2.3	4.7	>150	>150		
Ketoconazole					4.7	18.8

4. Conclusion

Two new Schiff bases *N*'-(3,5-dibromo-2-hydroxybenzylidene)-3-methylbenzohydrazideand *N*'-(4-chloro-2-hydroxybenzylidene)-4-fluorobenzohydrazide were prepared and utilized to assemble Cu^{II} ions to form three new copper complexes. The hydrazone coordinates to Cu(II) ions through phenolate oxygen, imino nitrogen, and carbonyl oxygen. The complexes have good antibacterial activity against *B. subtilis*, *S. aureus* and *E. coli*.

Supplementary Data

CCDC 2498992 (HL^1), 2498993 (**1**), 2482741 (**2**) and 2482742 (**3**) contain the supplementary crystallographic data for the complexes. These data can be obtained free of charge *via* http://www.ccdc.cam.ac.uk/conts/retrieving.html, or from the Cambridge Crystallographic Data Centre, 12 Union Road, Cambridge CB2 1EZ, UK; fax: (+44)1223-336-033; or e-mail: deposit@ccdc.cam.ac.uk.

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Povzetek

Halogenirane Schiffove baze imajo zanimive biološke aktivnosti. Sintetizirali smo dve novi Schiffovi bazi hidrazonskega tipa *N*'-(3,5-dibromo-2-hidroksibenziliden)-3-metilbenzohidrazid (HL^1) in *N*'-(4-kloro-2-hidroksibenziliden)-4-fluorobenzohidrazid (HL^2) z bromovimi, klorovimi in fluorovimi substituenti. Spojine reagirajo z različnimi bakrovimi solmi in tvorijo tri bakrove(II) komplekse z zanimivimi strukturami. Kompleksi so $[CuBrL^1]\cdot H_2O$ (**1**·H_2O), $[CuClL^2]\cdot CH_3OH$ (**2**·CH_3OH) in $[Cu_2Br_2L^2{}_2(CH_3OH)_2]$ (**3**). Hidrazona in trije bakrovi kompleksi so bili okarakterizirani s fizikalno-kemijskimi metodami, kot so elementarna analiza, infrardeča in elektronska spektroskopija. Prosta hidrazona sta bila okarakterizirani tudi z ^{1}H NMR spektroskopijo, strukture H_2L^1 in kompleksov so bile dodatno potrjene z monokristalno rentgensko analizo. Hidrazonski ligandi v kompleksih se vežejo na Cu ione prek fenolatnega kisika, imino dušika in karbonilnega kisika. Cu ion v kompleksu **1** je v kvadratni planarni koordinaciji, v kompleksu **2** v kvadratni piramidalni koordinaciji, v kompleksu **3** pa v oktaedrični koordinaciji. Kompleksi kažejo dobro antibakterijsko aktivnost na bakterijskih sevih *Bacillus subtilis*, *Staphylococcus aureus* in *Escherichia coli*, medtem ko imajo šibko aktivnost na glivičnem sevu *Candida albicans* in nobene aktivnosti na *Aspergillus niger*.