Dalton Transactions

An international journal of inorganic chemistry

Accepted Manuscript

This is an Accepted Manuscript, which has been through the Royal Society of Chemistry peer review process and has been accepted for publication.

Accepted Manuscripts are published online shortly after acceptance, before technical editing, formatting and proof reading. Using this free service, authors can make their results available to the community, in citable form, before we publish the edited article. We will replace this Accepted Manuscript with the edited and formatted Advance Article as soon as it is available.

You can find more information about Accepted Manuscripts in the Information for Authors.

Please note that technical editing may introduce minor changes to the text and/or graphics, which may alter content. The journal's standard Terms & Conditions and the Ethical guidelines still apply. In no event shall the Royal Society of Chemistry be held responsible for any errors or omissions in this Accepted Manuscript or any consequences arising from the use of any information it contains.

ROYAL SOCIETY OF CHEMISTRY

rsc.li/dalton

Synthesis of (3-nitro-2-oxo-benzaldehyde thiosemicarbazonato) - zinc(II) complexes : the position of nitro group in phenyl ring alters antimicrobial activity against *K. pneumoniae* 1, *S. typhimurium* 2, MRSA and *C. albicans*†

Tarlok S. Lobana,*[a] Shikha Indoria,[aΨ] Henna Sood,[c] Daljit S. Arora,[c] Manpreet Kaur[d] and Jerry P. Jasinski[d]

The basic purpose of this investigation was to explore the effect on antimicrobial activity of a nitro group at an ortho- versus para- position in the 2-hydroxy-phenyl ring of nitro-salicylaldehyde-N-substituted thiosemicarbazone (X-NO_2-stscH_2-N^1HR, X = 3 or 5; stsc- stands for salicyladehyde thiosemicarbazone) complexes with zinc. Reactions of zinc(II) acetate with 3-nitro-salicylaldehyde-N-substituted thiosemicarbazones (3-NO_2-stscH_2-N^1HR, **Chart 1**) and 2,2-bipyridine, or 1,10-phenanthroline as co-ligands, yielded complexes of stoichiometry, [Zn(3-NO_2-stsc-N^1HR)(N,N-L)] {L, R : bipy, H, **1**; Me, **2**; Et, **3**; Ph, **4;** phen, H, **5**; Me, **6**; Et, **7;** Ph, **8**}. The thio-ligands coordinate to the metal as dianions (deprotonation of –OH and -N^2H moieties) through O, N^3 and S donor atoms in distorted trigonal bipyramid geometry (**4, 5, 7**: τ = 0.718–0.576) or in distorted square pyramid geometry (**8** : τ = 0.349). ESI-mass spectrometry supported the formation of molecular ion peaks. Complexes displayed fluorescence with λ_{max} = 438-473 nm. It was found that these five-coordinated [Zn(3-NO_2-stsc-N^1HR)(N,N-L)] complexes showed high antimicrobial activity against methicillin resistant *S. aureus* (MRSA), *Klebsiella*

pneumoniae 1, *Salmonella typhimurium* 2 and *C. albicans* vis-à-vis that of 5-NO_2-stscH_2-N^{1}HR zinc complexes reported earlier. However, in comparison, the antimicrobial activity of 5-nitro complexes against *S. aureus* was high relative to 3-nitro complexes in the present case.

[a]*Department of Chemistry, Guru Nanak Dev University, Amritsar – 143 005, India*

[b]*Department of Microbiology, Guru Nanak Dev University, Amritsar-143 005, India*

[c]*Department of Chemistry, Keene State College, Keene NH 03435-2001, USA*

**E-mail: tarlokslobana@yahoo.co.in; Fax: +91-183-2258820.*

[Ψ]Present Address: Department of Chemistry, Kanya Maha Vidyalaya , Jalandhar-144004, India;

[†]Electronic supplementary information (ESI) available: CCDC numbers: 2020647, 2020648, 2020649 and 2020650 for complexes **4**, **5**, **7** and **8** respectively. For ESI and crystallographic data in CIF or other electronic format;

Introduction

Among the bio-metals (Co, Ni, Cu, Zn), the biochemistry of zinc(II) is important due to its role as a co-factor in a number of enzymes, namely, zinc proteinases, alkaline phosphatases, amino peptidases, carbonic anhydrases, histone deacetylases, etc. which are involved in various metabolic processes of plants, animals, viruses and bacteria.[1-8] In order to understand the role of zinc metal in biological processes, the coordination chemistry of this metal acquires importance especially with N,S-donor ligands.[9-14] In addition, there are efforts to develop metal based anti-

bacterial and anti-cancer drugs. In this respect, it is interesting to mention here that zinc(II) complexes of N,S-donor thiosemicarbazones (**Chart 1**) have shown antimicrobial,[15-18] anticancer (antitumour, antiproliferation, cytotoxicity)[19-22] and antioxidant activity.[23, 24] However, the anti-microbial studies reported in literature are preliminary and activity was also low.[15-18] The reported antimicrobial studies pertained to when R^1 at the C^2 atom of thiosemicarbazone was 2-thiophene, pyridine, furan, and 2-acetyl-butyrolactone, and R^2 was H or CH_3 group.[15-18]

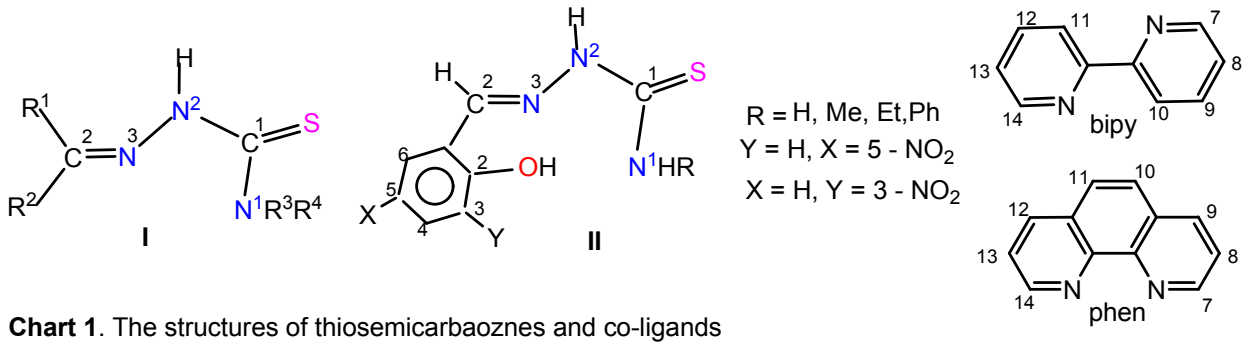

Chart 1. The structures of thiosemicarbaoznes and co-ligands

In view of our interest in coordination chemistry of thiosemicarbazones,[9,10] and biochemical importance of zinc(II), we thought to use O,N,S-donor salicylaldehyde based thiosemicarbazones and N,N-donor bipyriidnes / phenanthrolines as co-ligands, so as to obtain five coordinated complexes for studying their bio-activity. Thus we recently reported antimicrobial activity of complexes of this metal with 5-nitro-2-hydroxy-salicylaldehyde-N-substituted thiosemicarb - azones with 2,2′-bipyridines and 1, 10-phenanthrolines as co-ligands.[25,26] In the present paper, we report synthesis, structures and antimicrobial activity of zinc(II) complexes of 3-nitro-salicylaldehyde-N^1-substituted thiosemicarbazones (**Chart 1 :** R = H, Me, Et, Ph) with 2, 2′-bipyridine and 1,10 –phenanthroline as other organic ligands. The antimicrobial activity is investigated against methicillin resistant *Staphylococcus aureus* (MRSA - a clinical isolate of resistant bacteria), Gram positive bacteria, viz. *Staphylococcus aureus* (MTCC740), Gram

negative bacteria, *Klebsiella pneumoniae* 1 (MTCC109), *Salmonella typhimurium* 2 (MTCC1251), and a yeast, *Candida albicans* (MTCC227) {MTCC = Microbial Type Culture Collection}. The activity of 3- nitro-complexes is compared with 5-nitro complexes and the differences observed in two cases are highlighted.

Results and discussion

Synthesis and IR spectroscopy

Chart 2 lists zinc(II) complexes with 3-nitro-salicylaldehyde-N^1-substituted thiosemic – arbazones of stoichiometry, [Zn(3-NO_2-stsc-N^1HR)(L)],{L, R : bipy, H, **1**; Me, **2**; Et, **3**; Ph, **4;** phen, H, **5**; Me, **6**; Et, **7;** Ph, **8;** stsc - stands for salicylaldehyde thiosemicarbazone}. Here, the acetate anions of the zinc salt removed OH and hydrazinic -N^2H protons (Chart 2 for labeling, structure III) , and thus the thio-ligands coordinated to the Zn(II) metal center as O, N, S- donor atoms. The co-ligands, bipy and phen, are N,N-chelated to the metal center, forming five coordinated complexes (**1-8**). Complexes are soluble in dimethyl sulfoxide and partially soluble in dichloromethane, methanol and acetonitrile.

The IR spectra of these complexes revealed the absence of ν(O–H) and ν(N^2–H) bands, suggesting the loss of O-H and N^2-H protons (Chart 2. Structure III for labeling) and thus these thio-ligands are coordinated as di-anions, (3-NO_2-stsc-N^1HR)$^{2-}$. Further, the intense diagnostic ν(C-S) bands of the free ligands showed absorption in the region, 842-857 cm^{-1}, and in complexes, these bands shifted to the low energy region, 706 -775 (s) cm^{-1}. This large shift revealed a change of C=S bond to C-S bond in complexes with decreased pπ-pπ bond character in latter bonds. Thus this IR data supports that thio-ligands are coordinating as anionic ligands through O, N^3 and S donor atoms. The intense ν(N^1–H) bands of the –C^1(=S)–N^1HR moiety of

the free ligands (Chart 2. Structure III), which occurred in the region, 3351-3452s cm^{-1}, changed to the broad bands in the range, 3400- 3450 cm^{-1}, in these complexes. The medium to strong IR bands of the free ligands, due to the ν(N–O) stretching, appeared in the regions, 1531-1550s and 1353-1385m-s, and in the complexes, these bands shifted to the regions, 1508-1604m-s and 1316-1354m-s (See ESI and experimental for details; b-broad, m-medium, s-strong).

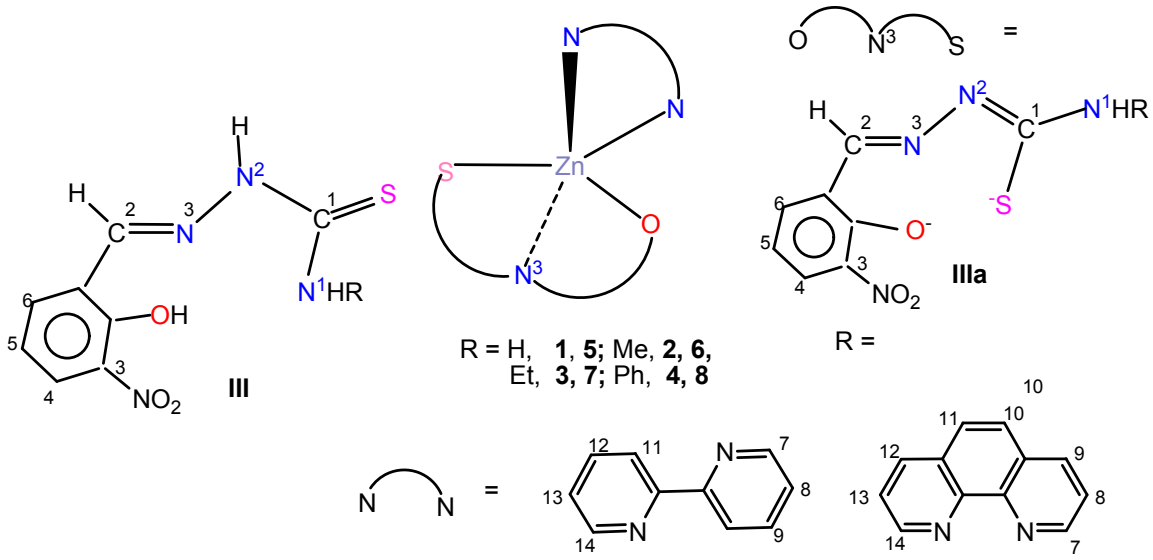

Chart 2..Representation of zinc derivatives of 3-nitro-salicylaldehyde thiosemicarbazones (R =H, Me, Et, Ph)

NMR, Electronic absorption and Fluorescence spectroscopy

The uncoordinated thio-ligands, 3-NO_2-stscH_2-N^1HR (R = H, Me, Et, Ph), showed bands due to –OH and –N^2(H)- moieties (Chart 2. Structure III) in the regions, 10.99 to 11.53 ppm and 8.47-10.21 ppm respectively. These signals disappeared in the complexes (experimental section), supporting that the thio-ligands lost both the protons in complex formation and coordinated to the metal center as di-anions (see ESI detailed proton NMR data of free ligands, bipy / phen co-ligands). In complexes, **1-8**, the ^{1}H NMR signals due to the coordinated thio-ligands (e.g. C^2H,

N^1H, N^1CH_3, N^1CH_2-, N^1CH_3- and N^1Ph protons), and 2,2′-bipyridine and 1,10-phenanthroline co-ligands are listed in the experimental section, which are at different positions relative to those of the uncoordinated ligands (ESI), supporting the formation of complexes. The proton NMR signals due to the aromatic ring of 3-NO_2-C_6H_4- moiety of the thio-ligands deserve special comment being close to the coordinating O and N^3 donor atoms. Here C^4H, C^5H and C^6H protons are under different chemical environments, and thus each type of these hydrogens must show a doublet of doublet (C^4H coupling with C^5H should give doublet and further coupling to C^6H proton should give a doublet of doublet and same is expected for C^5H and C^6H protons). The coupling, if not resolved, will give rise to less than the expected signals or mutiplets. Thus the free ligands, 3-NO_2-stscH_2-N^1HR, showed a doublet each for C^4H +C^6H protons with the R group being H, Me or Ph at N^1 atom, and a multiplet for the ligand with R = Et group. Further, the C^5H proton showed a doublet each for the ligands with R = H, Et, or Ph groups and a multiplet for R= Me (ESI). In the complexes, the C^4H +C^6H protons appeared as a singlet (**2**, **4**, **8**), a doublet (**3**, **5**) or a multiplet (**1**, **6**, **7**), while the C^5H proton appeared as a singlet (**2-7**), a doublet (**8**), or a triplet (**1**). Therefore, these data support that in no case we got an expected pattern of a doublet of doublet in uncoordinated thio-ligands, or in complexes. From the above observations, it is inferred here that the chemical environment difference between C^4H, C^5H and C^6H protons is very small and resolution has not occurred, rather only an unresolved singlet, a doublet, or an overlapping multiplet were obtained.

The electronic absorption spectra of a 10^{-4} M solution of these complexes are shown in Fig. 1, which occur in the UV–visible region. Broadly, the absorption bands occur in the two regions: 265-325 nm region, assigned to the $\pi\rightarrow\pi^*$ transitions centered on phenyl / pyridyl rings of ligands, and 360-373 nm region, assigned to the $n\rightarrow\pi^*$ transitions attributed to the azomethine

(C=N) / thiouride (C=S) groups of thio-ligands. The bands in the region 408-428 nm arise, probably, due to the MLCT electronic absorption bands, involving metal to π* orbitals of bipy/ phen ligands.[27] Further, the complexes exhibited fluorescence in the range, 370-560 nm, with λ_{max} at 438-473 nm, corresponding to the excitation wavelength, λ = 340 nm (**Fig. 2**). The origin of fluorescence appears to be linked to {Zn(N,N-L} moiety (N, N-L = bipy, phen) involving the intra-ligand transitions.[25,26] The fluorescence bands are associated with the vibrational fine structures.

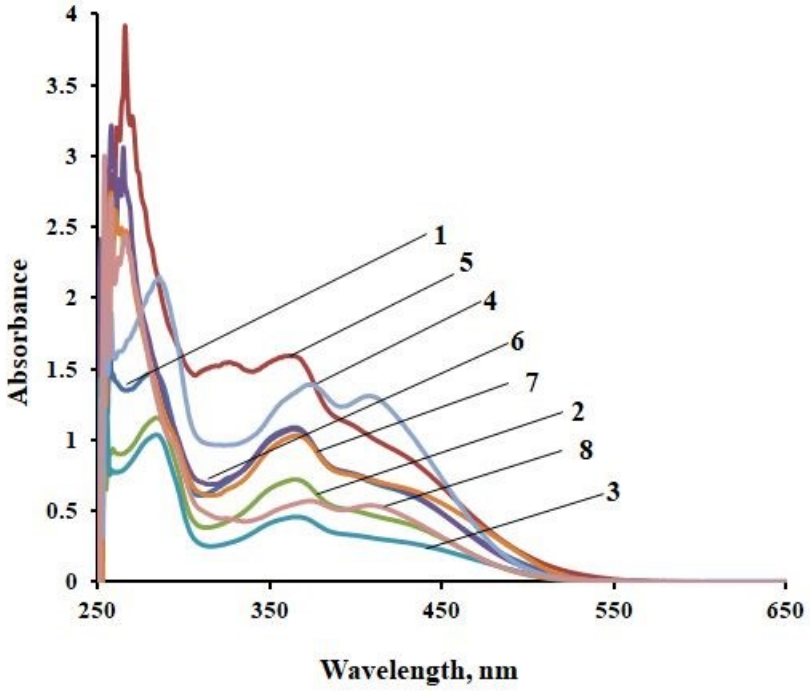

Fig. 1. The electronic absorption spectra of complexes, **1-8**

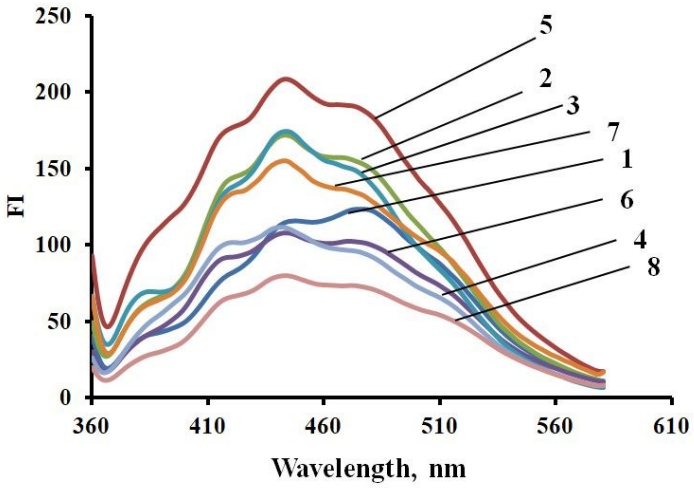

Fig. 2. Fluorescence spectra of zinc(II) complexes, **1-8**

Molecular structures

The crystal structure of complexes, [Zn(3-NO_2-stsc-N^1HPh)(bipy)] **4**, [Zn(3-NO_2-stsc-N^1H_2)(phen)] **5**, [Zn(3-NO_2-stsc-N^1HEt)(phen)] **7,** and [Zn(3-NO_2-stsc-N^1HPh)(phen)] **8**, have been solved using single crystal X-ray crystallography. Other complexes did not form suitable crystals for X-ray crystallography. The crystals of **4** and **7** are orthorhombic and triclinic respectively, with the corresponding space groups, Pbca and P-1, while complexes **5** and **8** formed monoclinic crystals with their respective space groups, $P2_1/n$ and $P2_1/c$. Table 1 contains crystal data, while Table 2 has selected bond parameters. The molecular structure of complex **4** (Ph, bipy), shows that zinc is bonded to an oxygen, azomethine nitrogen (N^3) and sulfur donor atoms of the (3-NO_2-stsc-N^1HPh)$^{2-}$anion. Zinc is further bonded to both the nitrogen atoms of the bipy ligand forming a N, N-chelate (**Fig.** 3). The coordination patterns of other structurally characterized complexes, **5** (H, phen), **7** (Et, phen) and **8** (Ph, phen), are similar with the only difference being of 1,10-phenanthroline as a co-ligand (**Figs**. 4-6). Complex **7** (Et, phen) has two

crystallographically independent molecules (I and II) in the asymmetric unit. Other complexes (**1-3, 6**) are suggested to have similar structures.

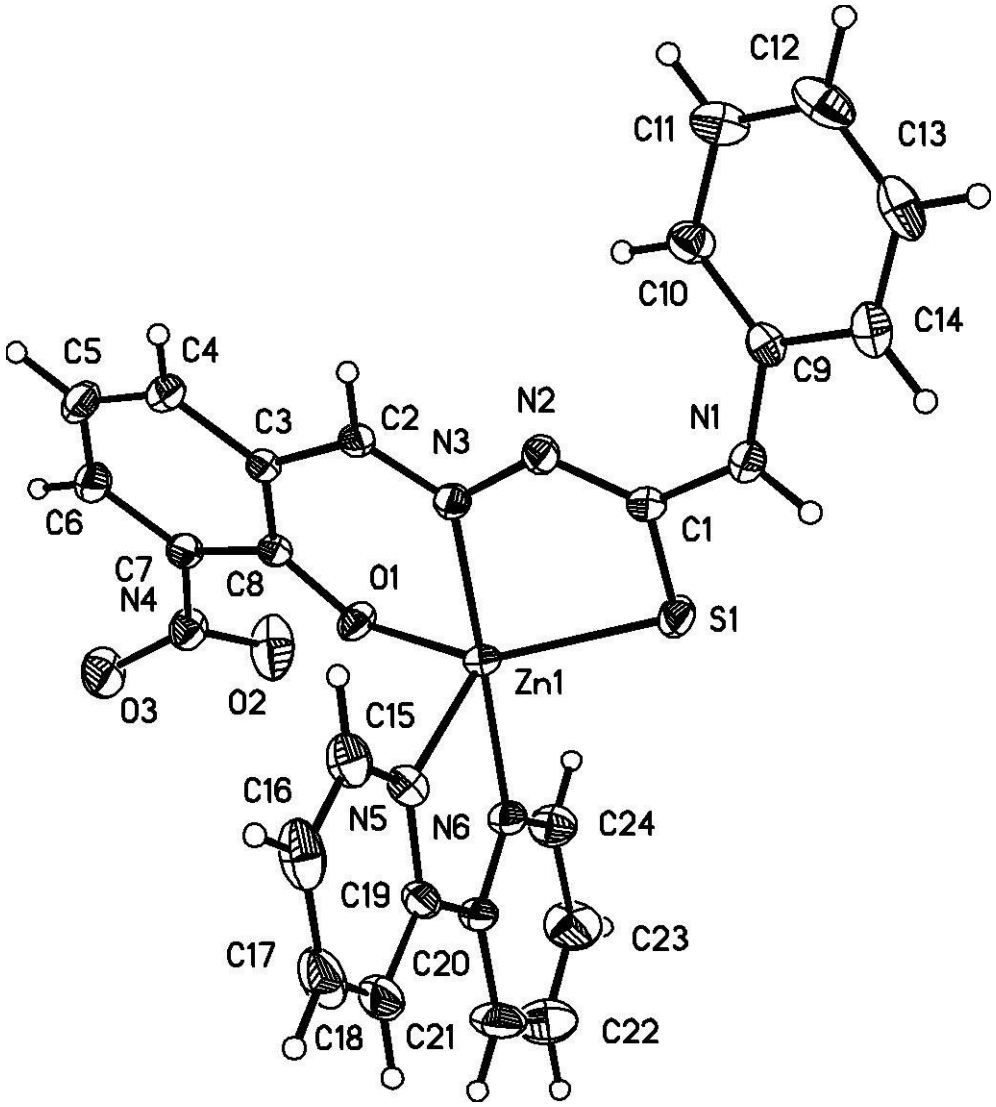

Fig. 3. Molecular structure of [Zn(3-NO_2-stsc-N^1HPh)(N,N-bipy)] **4**

The zinc-donor atom bond distances fall in the narrow ranges: Zn-O, 1.959 - 2.012; Zn-S, 2.317 - 2.356; Zn - N_{azo}, 2.067 - 2.112 and Zn - $N_{(bipy/phen)}$, 2.098 - 2.159 Å, which are normal and analogous to those reported in the literature.[25, 26] The O-Zn-N_{azo} bond angles of six-membered

chelate rings, fall in the range, 82-89°, while S-Zn-N_{azo} and N-Zn-$N_{(bipy/phen)}$ bond angles of five-membered chelate rings, fall in the ranges, 80-83 and 77-80°, respectively. Further, the largest bond angles, N_{azo}-Zn-$N_{A(bipy/phen)}$ fall in the range, 172-179°, while O-Zn-S and N_{azo}-Zn-$N_{B(bipy/phen)}$ bond angles fall in the next lower ranges, 134-152° and 98-108°, respectively. It is added here that the distortion value of the coordination polyhedron, $\tau = (\beta-\alpha)/60$, is evaluated by the two largest bond angles (α, β) in five coordination geometry (τ = 1 for ideal trigonal bipyramid and 0 for square pyramidal environment).[28] The τ-parameters of complexes **4**, **5**, **7** and **8** varied as follows: 0.593 (**4**), 0.718 (**5**, Molecule I), 0.576 (**5**, Molecule II), 0.639 (**7**) and 0.349 (**8**), respectively. The above τ-values suggest distorted trigonal bipyramid geometry for **4**, **5** and **7** complexes, and a distorted square pyramid geometry for complex **8**. The C-S bond distances fall in the close range of 1.742 – 1.757 Å. The C-S bond single bond distance is 1.81 Å and the C=S double bond distance is 1.69 Å. The C-S bonds in the present complexes have a bond distance in the middle of two extremes.[29] It shows weakening of pπ-pπ bonds in complexes and it further confirms that a thio-ligand binds to a Cu metal center as anion.

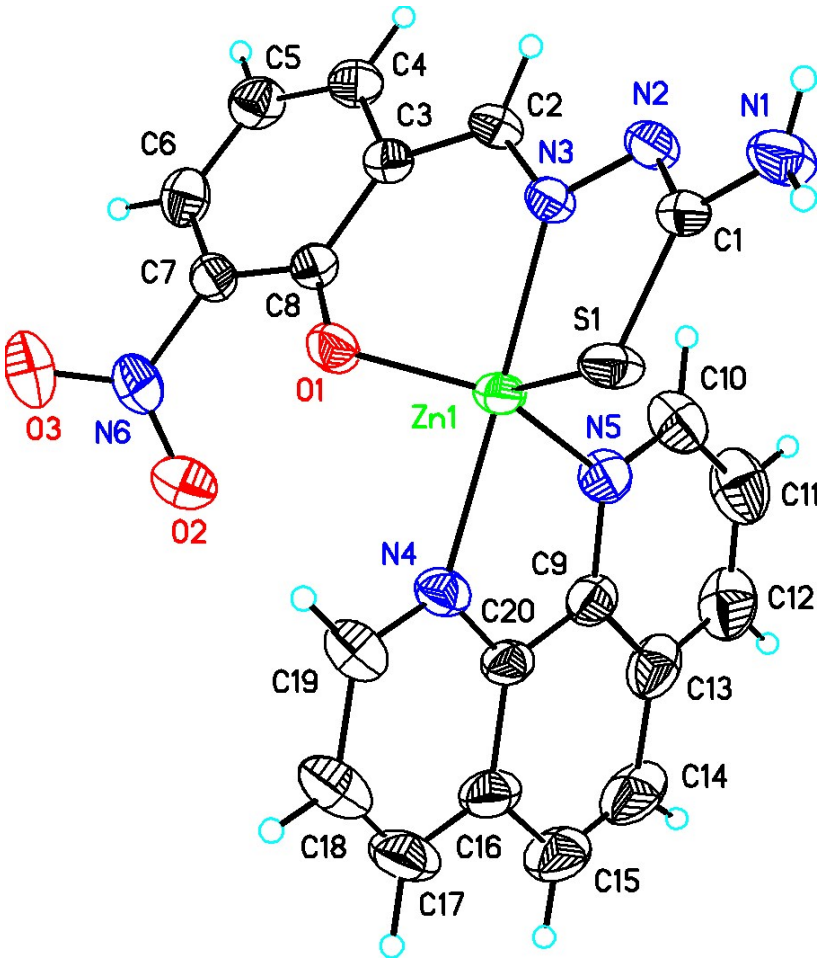

Fig. 4. Molecular structure of [Zn(3-NO_2-stsc-N^1H_2)(N,N-phen)] **5**

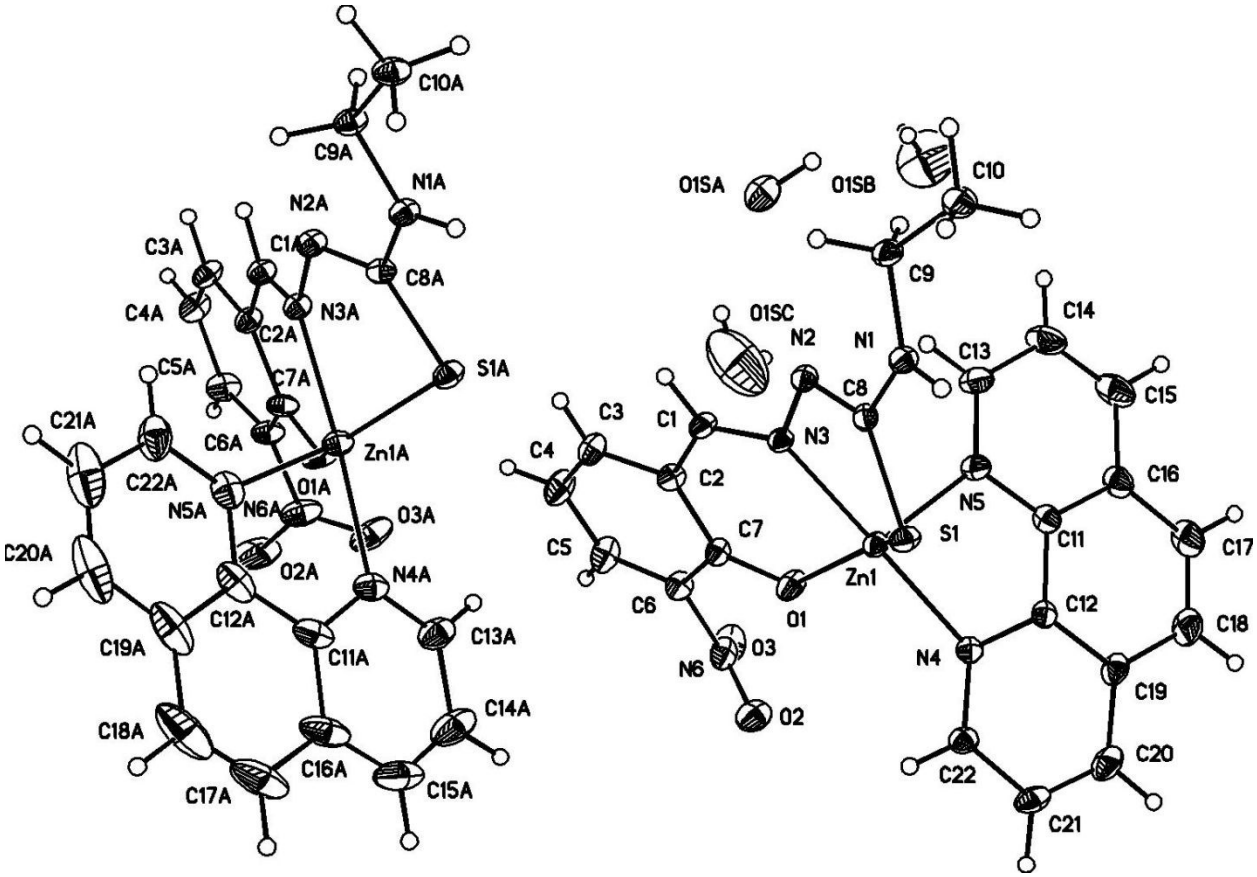

Fig. 5. Molecular structure of [Zn(3-NO_2-stsc-N^1HEt)(N,N-phen)] **7**

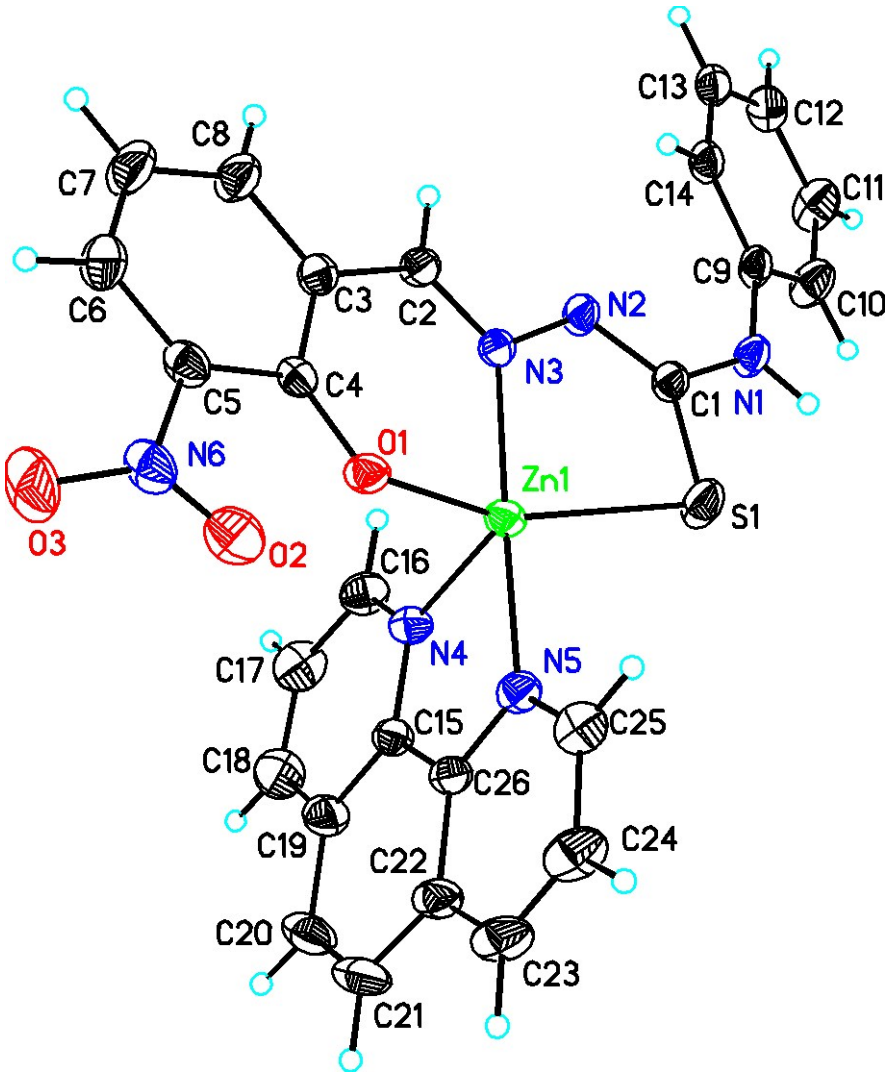

Fig. 6. Molecular structure of [Zn(3-NO_2-stsc-N^1HPh)(N,N-phen)] **8**

Table 1 Crystallographic data for complexes **4**, **5**, **7** and **8**

	4(Ph, bipy)	**5** (H, phen)	**7** (Et, phen)	**8**(Ph, phen)
CCDC	2020647	2020648	2020649	2020650
Empirical formula	$C_{24}H_{18}N_6O_3SZn$	$C_{20}H_{14}N_6O_3SZn$	$2(C_{22}H_{18}N_6O_3SZn)\cdot 3(H_2O)$	$C_{26}H_{18}N_6O_3SZn$
fw	535.87	483.80	1077.75	559.89
T(K)	173(2)	173(2)	173(2)	173(2)
λ (Å)	1.54184	1.54184	0.71073	0.71073
Crystal system	Orthorhombic	Monoclinic	Triclinic	Monoclinic
Space group	P b c a	$P2_1/n$	P-1	$P2_1/c$
Unit cell dimensions				
a(Å)	9.63257(14)	9.4903(2)	12.1163(8)	9.5596(7)
b(Å)	16.0028(3)	13.9811(4)	13.7570(10)	30.0912(14)
c(Å)	31.8147(5)	15.4215(4)	17.8138(9)	9.0688(5)
α(°)	90	90	109.265(6)	90
β(°)	90	104.862(3)	94.725(5)	112.729(8)
γ(°)	90	90	113.463(7)	90
V(Å^3)	4904.17(13)	1977.74(10)	2491.7(3)	2406.1(3)
Z	8	4	2	4
D_{calcd}(g cm^{-3})	1.452	1.625	1.436	1.546
μ(mm^{-1})	2.509	3.037	1.111	1.150
F(000)	2192	984	1108	1144
GOF	1.082	1.037	1.099	1.163
Reflections collected	39891	6846	34122	12495
Unique reflections	4723, (R_{int} = 0.0523)	3760, (R_{int}= 0.0443)	16383, (R_{int} = 0.0463)	5292 (R_{int} = 0.0352)
Data/restraints/ parameters	4723 / 0/ 316	3760 / 0 /297	16383/ 0 / 633	5292 / 0 / 334
Reflens.with [I>2σ(I)]	4257	3299	9944	4288
R Indices				
R_1	0.0418	0.0425	0.0665	0.0567
wR_2	0.1067	0.1092	0.1697	0.1147
R indices (all data)				
R_1	0.0458	0.0473	0.1163	0.0738
wR_2	0.1089	0.1159	0.1905	0.1246

Largest diff. Peak and hole	0.905 and -0.340 e.Å^{-3}	0.409 and -0.530 e.Å^{-3}	0.971 and -0.806 e.Å^{-3}	0.910 and -0.676 e.Å^{-3}

Table 2 Important bond distances (Å) and bond angles (°) of complexes **4**, **5**, 7 and **8**

	4(Ph, bipy)	**5**(H, phen)	**7**(Et, phen)		**8** (Ph, phen)
			I	**II**	
Zn–O	1.972(2)	1.959(2)	1.966(2)	1.974(2)	2.018(2)
Zn–$N_{(azo)}$	2.0891(19)	2.112(2)	2.086(3)	2.090(3)	2.065(3)
Zn–S	2.3245(7)	2.317(1)	2.355(1)	2.353(1)	2.356(1)
Zn–N	2.099(2), 2.152(2)	2.115(2), 2.159(2)	2.098(3), 2.136(3)	2.112(3), 2.156(3)	2.150(3) 2.099(3)
C-S	1.745(2)	1.742(2)	1.757(3)	1.749(3)	1.743(4)
O-Zn-N_{azo}	88.43(7)	87.32(7)	89.06(10)	88.70(10)	87.86(9)
S-Zn-N_{azo}	82.44(6)	82.06(5)	80.72(8)	81.02(8)	82.51(8)
O-Zn-S	143.03(6)	134.34(6)	144.31(9)	137.99(9)	151.30(8)
N_{azo}-Zn- N_B	101.65(8)	101.33(8)	100.92(11)	98.32(13)	107.85(11)
N_{azo}-Zn-N_A	178.63(8)	177.46(7)	178.88(10)	176.37(12)	172.21(11)
N_A-Zn- N_B	77.27(8)	77.89(8)	79.42(11)	78.21(14)	79.32(11)
τ value	0.593	0.718	0.576	0.639	0.349

N_A = N atom of bipy/phen with largest bond angle; N_B = N atom of bipy/phen with smaller angle; I = Molecule **I**, II = Molecule **II**.

ESI-mass studies

ESI-mass spectra of complexes **1-3**, **5** and **8** have been recorded. These complexes showed intense molecular ions, $[M+H]^+$, which are listed here : **1**, calc. for $[Zn(3\text{-}NO_2\text{-}stsc\text{-}N^1H_2)(bipy) + H]^+$ 459.02, obsd. 459.01; **2**.calc. for $[Zn(3\text{-}NO_2\text{-}stsc\text{-}N^1HMe)(bipy) + H]^+$ 473.03, obsd. 472.98; **3**. Calc. for $[Zn(3\text{-}NO_2\text{-}stsc\text{-}N^1HEt)(bipy) + H]^+.H_2O$ 505.06, obsd. 505.02. Complexes

5 and **8** also showed molecular ions; **5**. calcd for $[Zn(3\text{-}NO_2\text{-}stsc\text{-}N^1H_2)(phen)+H]^+$ 483.02, obsd. 482.99 and finally, **8.** calc. for $[Zn(3\text{-}NO_2\text{-}stsc\text{-}N^1HEt)(phen) + H]^+$ 559.05; obsd. m/z = 559.01. Figs.7 and 8 depict the mass spectral peaks of molecular ions of two representative complexes **1** and **8** (see ESI for more details).

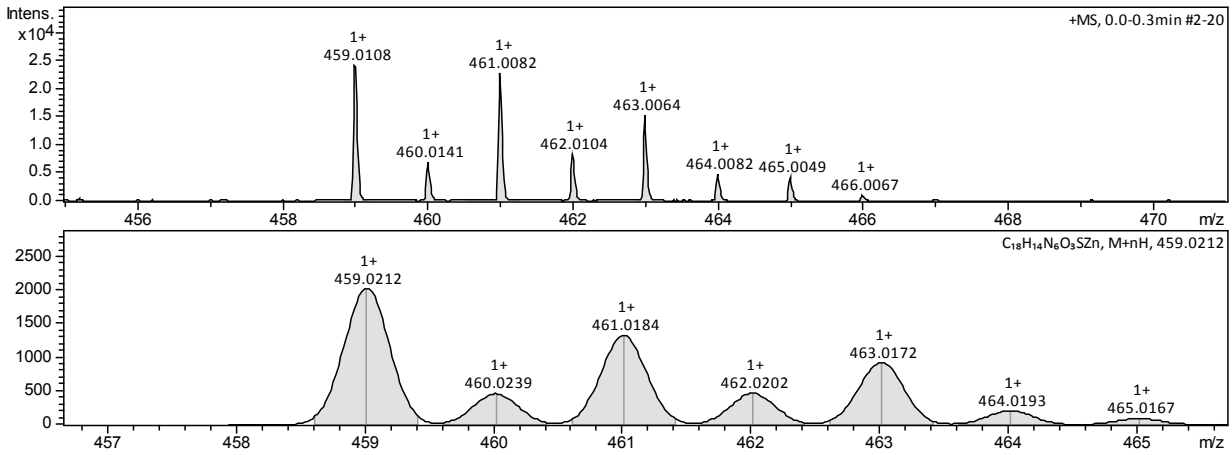

Fig. 7. ESI-mass peak due to molecular ion $[Zn(3\text{-}NO_2\text{-}stsc\text{-}N^1H_2)(bipy)+H]^+$ with isotopic pattern (complex **1**).

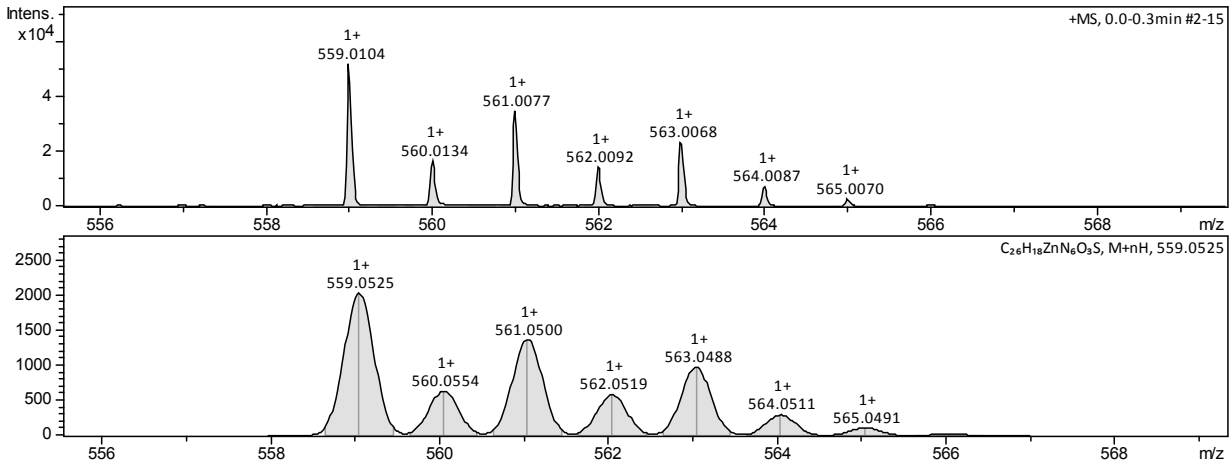

Fig. 8. ESI-mass peak due to molecular ion $[Zn(3\text{-}NO_2\text{-}stsc\text{-}N^1HPh)(phen)+H]^+\cdot H_2O$ with isotopic pattern (complex **8**).

Antimicrobial activity

Table 3 presents minimum inhibitory concentration (MIC, μg mL^{-1}) of these zinc(II) complexes, $[Zn(3\text{-}NO_2\text{-}stsc\text{-}N^1HR)(L)]$ {L, R : bipy, H, **1**; Me, **2**; Et, **3**; Ph, **4;** phen, H, **5**; Me, **6**; Et, **7;** Ph, **8**}. Among these, the complexes **2**, **3**, **4** and **7** showed high antimicrobial activity (MIC, 10 μg mL^{-1}) against methicillin resistant *S. aureus* (MRSA), and this activity is rather same as that of the reference drug, gentamicin (MIC = 10 μg mL^{-1}).[26,30] In literature, it was noted that among several complexes investigated, only selected Cu^{II}/Zn^{II} complexes with 5-methoxy-, or 5-nitro-salicylaldehyde thiosemicarbazones, showed moderate to high antimicrobial activity against MRSA at low MIC values, in the range, 5-7 μg mL^{-1}, [26, 31] at which gentamicin was inactive.[25, 31, 32] As regards the *Staphylococcus aureus* microorganism, only complex **2** showed the highest activity with MIC value of 7 μg mL^{-1}, which was, however, less than that of the reference drug (0.5 μg mL^{-1}, Table 3). Other complexes **3-5** and **7** showed the next lower activity (each complex with MIC value of 50 μg mL^{-1}).

Table 3 Antimicrobial data for zinc complexes **1-8** (minimum inhibitory concentration, MIC, μg mL^{-1}) [a-d]

Complex No.	*MRSA* MIC	*S. aureus* MIC	*K. pneumoniae 1* MIC	*S. typhimurium 2* MIC	*C. albicans* MIC
1 (H, bipy)	750	1000	500	NA	750
2 (Me, bipy)	10	7	50	5	7

3 (Et, bipy)	10	50	500	750	7
4 (Ph, bipy)	10	50	500	50	50
5 (H, phen)	500	50	500	ND	5
6 (Me, phen)	50	1000	500	NA	1000
7 (Et, phen)	10	50	7	1000	7
8 (Ph, phen)	50	750	500	ND	1000
Gentamicin[b]	10	0.5	0.3	1	-
Amphotericin[c]	-	-	-	-	0.1

[a]Studies were made in 30% DMSO, at which there is no antimicrobial actvity. [b]Gentamicin was used as positive control against bacteria {*MRSA, S. aureus, S. typhimurium* 2*, K.pneumoniae* 1) and [c]Amphotericin acts as a positive control against yeast (*C. albicans*); [d] see ESI for zone of inhibition of complexes and ligands.

Among the complexes tested, only two complexes showed high activity (7 μg mL^{-1} , complex **7**; 50 μg mL^{-1}, complex **2)** against *Klebsiella pneumonia* 1**.** However, this activity was less in comparison to that of the reference drug (MIC, 0.3 μg mL^{-1}). As regards the antimicrobial activity against *Salmonella typhimurium* 2, only complexes **2** and **4** showed high activity (MIC, 5 μg mL^{-1} complex **2**, MIC, 50 μg mL^{-1} complex **4**), while the activity of other complexes was low. However, this activity of complexes **2** and **4** is less than that of the reference drug (MIC, 1 μg mL^{-1}). Other complexes either showed very low activity (**3** and **7**), or were inactive (**1**, **5**, **6** and **8**). In respect of *Candida albicans*, complexes **2**, **3**, **5** and **7** showed high activity (MIC values of 5-7 μg mL^{-1}) relative to that of other complexes (**1, 4, 6** and **8**). However, this activity is less in comparison to that of the reference compound, amphotericin (MIC, 0.1 μg mL^{-1}).

The most active complexes, namely, **2** (Me, bipy), **3** (Et, bipy), **5** (H, phen) and **7** (Et, phen) were further studied for their *in vitro* cell viability. This was evaluated by the colorimetric MTT assay method {MTT = 3-[(4, 5-dimethylthiazol-2-yl)-2, 5-diphenyl]tetrazolium bromide}.[33] The method is based on the reduction of a water soluble yellow substrate MTT, by mitochondrial succinate dehydrogenase enzyme present in blood cells, into an insoluble purple formazan product. These formazan crystals were then dissolved in dimethyl sulfoxide (Control diluent, 30% DMSO) followed by the measurement of their absorbance at λ_{max} = 595 nm. However, the cells exposed to complexes did not show measurable absorbance, thus ruling out the possibility of cell viability. It is concluded that these complexes have bactericidal / fungicidal properties, as was found with the analogous zinc(II) complexes of 5-nitro-salicyclaldehyde-N[1]-substituted thiosemicarbazones with 2,2′-bipyridine / 1,10-phenanthroline as co-ligands.[25]

A comparison of antimicrobial data with analogous reported zinc complexes

In this section, an effort is made to correlate the effect on bio-activity of complexes with shifting of the nitro substituent in the 2-hydroxyphenyl ring from a para (5) to an ortho (3) position, relative to the 2-hydroxy substituent in the phenyl ring (see Chart 3). Table 4 shows a comparative data of the antimicrobial activity of zinc(II) complexes with 3-nitro- and 5-nitro-salicylaldehyde-N-substituted thiosemicarbazones. In this comparison, the highest activity of these complexes against MRSA*, S. aureus, K. pneumoniae 1* ***and*** *S. typhimurium 2* bacterial strains and one yeast *C. albicans* is considered. Each of 3-nitro complexes (**2**, **3**, **4**, **7**) showed an activity of 10 μg mL^{-1}against MRSA, while each of 5-nitro complexes (**2-5, 7, 8**) showed low activity of 50 μg mL^{-1}. In respect of *K. pneumoniae 1,* complex **7** showed high activity (7 μg mL^{-1}) versus low activity (50 μg mL^{-1}) shown by 5-nitro

complexes (**2, 3, 7**). Similarly, against *S. typhimurium 2* and *C. albicans,* 3-nitro complexes are more active than the 5-nitro-complexes. However, against *S. aureus,* several 5-nitro- complexes (**2 - 5, 7, 8)** showed high activity (0.5 μg mL^{-1}) versus low activity (7 μg mL^{-1}) shown by a 3-nitro-complex (**2**).

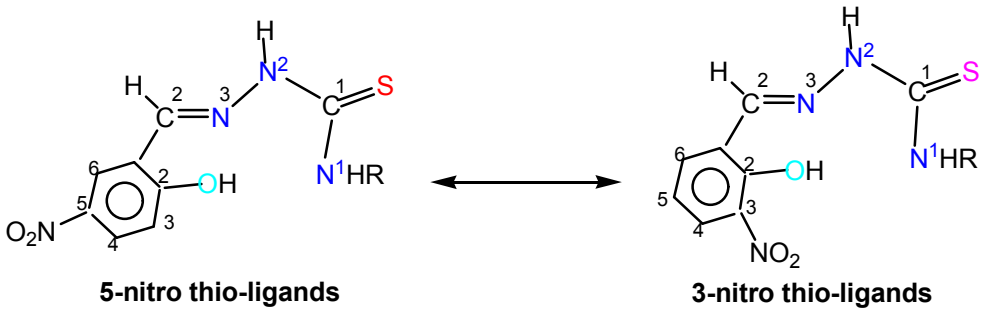

Chart 3. The 3- and 5-nitro-salicylaldehyde-N^1-substituted thiosemicarbazones

Table 4 A comparative antimicrobial data of 3-nitro- and 5-nitro- zinc complexes with salicylaldehyde-N^1-substituted thiosemicarbazones

	3-Nitro complexes **[This work]**		5-Nitro complexes[a] [Ref. 25]	
Microorganism	Complex number	MIC, μg mL^{-1}	Complex number	MIC, μg mL^{-1}
MRSA	**2, 3, 4, 7**	10	**2 - 5,7, 8**	50
S. aureus	**2**	7	**2 - 5, 7, 8**	0.5
K. pneumoniae 1	**7**	7	**2, 3, 7**	50

S. typhimurium2	**2**	5	**2, 3, 5, 7, 8**	10
C. albicans	**2, 3, 5, 7**	5-7	**8**	10

[a]5-Nitro-complexes are numbered in brick red colored : Zn(5-NO_2-stsc-N^1HR)(N,N-L)] {L, R : bipy, H, **1;** Me, **2**; Et, **3** ; Ph, **4 ;** phen, H′, **5** ; Me, **6** ; Et, **7;** Ph, **8**}.

Conclusion

A series of five coordinated zinc(II) complexes of stoichiometry, [Zn(3-NO_2-stsc-N^1HR)(N,N-L)] (R = H, Me, Et, Ph; L= bipy; **1-4; phen, 5-8)**), have been synthesized using 3-nitro-salicylaldehyde-N^1-substituted-thiosemicarbazones with 2,2-bipyridine and 1,10-phenanthroline as other organic ligands. The x-ray crystallography revealed distorted trigonal bipyramidal, or square pyramidal geometry of complexes studied. The variation in position of nitro group from 5-nitro- to 3-nitro in phenyl ring of 2-hydroxyphenyl- moiety at C^2-position of the thio-ligands though did not affect the geometry of complexes, but significantly changed antimicrobial activity of bacterial and fungi strains investigated. The 3-nitro-complexes showed high activity against methicillin resistant *S. aureus,* Gram negative bacteria, *K. pneumoniae* 1, *Salmonella typhimurium* 2 and the yeast *C. albicans* vis-à-vis the 5-nitro-complexes reported. In contrast 5-nitro complexes were more active against *S. aureus*. This is an interesting outcome of the present study. It revealed that the design of a ligand is important to influence the bio-activity of complexes. However, the complexes did not show any measurable cell viability, and thus these zinc complexes have bactericidal / fungicidal properties. The variation of co-ligands could

be another perturbation to alter bio-activity of complexes with the aim to investigate bio-compatible materials.

Experimental

Materials and techniques

Zinc(II) acetate dhydrate, thiosemicarbazide, N-methyl thiosemicarbazide, N-ethyl thiosemicarbazide, N-phenyl thiosemicarbazide, 3-nitro-salicylaldehyde, 2,2-bipyridine (bipy) and 1,10-phenanthroline (phen) were procured from Aldrich Sigma Ltd. The thio-ligands were synthesized according to the reported procedures.[34-36] Elemental analysis (C, H, and N) were carried out with a thermoelectron FLASHEA1112 analyzer. Melting points were determined with a Gallenkamp electrically heated apparatus. The UV-visible spectra of 10^{-3}-10^{-4} M solutions of the compounds were recorded in dimethyl sulfoxide (dmso) with the help of a UV-1601 PC Shimadzu spectrophotometer. Fluorescence spectra of complexes (10^{-4} M) were recorded with a Varian Cary Eclipse Fluorescence spectrophotometer. The IR spectra of the compounds were recorded in the 4000 – 450 cm^{-1} region with a Perkin Elmer FT-IR Spectrometer by making their KBr pellets. Proton NMR spectra of compounds were recorded on a Bruker Avance 500MHz NMR spectrometer in a mixture of $CDCl_3$ and dmso (9 : 1 : : v/v) with TMS as the internal reference. The ESI-mass spectra were recorded in DMSO or $CHCl_3$ solvents using a Bruker Daltonik LS-MS high resolution micro TOF-Q II 10356 spectrometer.

Synthesis of zinc(II) complexes

[Zn(3-NO_2-stsc-N^1H_2)(bipy)] (1) . To an orange solution of thio-ligand, 3-NO_2-stscH_2-N^1H_2 (0.027 g, 0.11 m mol) in acetonitrile (10 mL) was added solid $Zn(OAc)_2 \cdot 2H_2O$ (0.025 g, 0.11 m mol). The reaction mixture was stirred for 15 minutes which yielded a pale yellow precipitate.

To the precipitate, a solution of bipyridine (0.017 g, 0.11 mmol) in dichloromethane (10 mL) was added and the contents were again stirred for 15 min. A clear dark orange solution formed was allowed to evaporate at room temperature which yielded an orange compound. Yield, 0.037 g, 71%, mp 219-221°C. Elemental analysis for $C_{18}H_{14}N_6O_3SZn$:calcd C 45.25; H 3.38; N 17.59; S 6.71(%); found: C 45.38; H 3.13; N 17.64; S 6.83. Main IR Bnads (KBr, cm^{-1}): ν(N^1–H) 3450sb; ν(C–H) 2966w, 2932w, 2808w; ν(C=N) + ν(C=C) + δ(N−H) + ν(N=O) 1600s; δ(C−H) 1470w, 1446w; 1383w; δ(N=O) 1354s; 1238w, 1154w, 1143m, 954w; ν(C–S) 766s; 708m, 641 w, 587w, 425w, 404w. Electronic absorption spectrum (DMSO, λ_{max} /nm, ε / L mol^{-1} cm^{-1}): [10^{-4} M] 408 (7.14 x 10^3); 373 (1.39 x 10^4), 286 (2.14 x 10^4). Fluorescence spectrum: (λ_{max}^{em} = 442; λ_{max}^{ex} = 340 nm). ^{1}H NMR (δ, ppm; $CDCl_3$: DMSO; 9:1): δ = 8.60 (2H, s, C^7H_{bipy} + $C^{14}H_{bipy}$), 8.50 (1H, s, C^2H), 8.29 (2H, d, $C^{10}H_{bipy}$ + $C^{11}H_{bipy}$), 7.96 (2H, m, C^4H + C^6H), 7.99 (2H, s, C^9H_{bipy} + $C^{12}H_{bipy}$), 7.64 (2H, s, C^8H_{bipy} + $C^{13}H_{bipy}$), 7.54 (2H, d, N^1H_2), 6.37 (1H, t, C^5H). ESI mass data: calcd for $C_{18}H_{14}N_6O_3SZn$, $[Zn(3\text{-}NO_2\text{-}stsc\text{-}N^1H_2)(bipy)+H]^+$, m/z = calcd 459.02, obsd. 459.01. Complexes **2-8** were similarly prepared.

[Zn(3-NO_2-stsc-N^1HMe)(bipy)] (2). Yield, 0.043 g, 78%, mp 204-206°C. Elemental analysis for $C_{19}H_{16}N_6O_3SZn$: calcd C 48.16; H 3.40; N 17.74; S 6.77(%); found: C 47.98; H 3.69; N 17.35; S 6.78%. Main IR Bands (KBr, cm^{-1}): ν(N^1–H) 3420sb; ν(C–H) 2974w, 2937w, 2812m; ν(C=N) + ν(C=C) + δ(N−H) 1604s; ν(N=O) 1508m, δ(C−H) 1446m; 1387m; δ(N=O) 1354s; 1270s, 1242s, 1162w, 966s, 879m; ν(C–S) 766s; 742s, 658m, 649s, 596m, 521w, 438m, 412 m. Electronic absorption spectrum (DMSO, λ_{max} /nm, ε / L mol^{-1} cm^{-1}): [10^{-4} M] 418 (4.49 x 10^3), 363 (7.24 x 10^3), 287 (1.25 x 10^4). Fluorescence spectrum: (λ_{max}^{em} = 443 nm; λ_{max}^{ex} = 340 nm). ^{1}H NMR (δ, ppm; $CDCl_3$: DMSO; 9:1): δ = 8.58 (2H, s, C^7H_{bipy} + $C^{14}H_{bipy}$); 8.39 (H, d, C^2H), 8.29 (2H, s, $C^{10}H_{bipy}$ + $C^{11}H_{bipy}$), 8.01 (2H, s, C^4H+C^6H), 7.92 (1H, s, N^1H),

7.53 (2H, m, C^9H_{bipy} + $C^{12}H_{bipy}$), 7.50 (3H, s, C^8H_{bipy} + $C^{13}H_{bipy}$ + N^1H), 6.35 (1H, s, C^5H), 3.14 (3H, d, CH_3N^1)). ESI mass data: calcd for $C_{19}H_{16}N_6O_3SZn$, [Zn(3-NO_2-stsc-N^1HMe)(bipy)+H]$^+$ **,** m/z = calcd 473.03; obsd. 472.98.

[Zn(3-NO_2-stsc-N^1HEt)(bipy)].CH_3CN (3). Yield, 0.042 g, 76%, mp 190-192°C. Elemental for $C_{20}H_{18}N_6O_3SZn$: calcd C 49.24; H 3.72; N 17.23, S 6.57% ;found: C 49.10; H 3.49; N 17.41, S 6.72%. Main IR Bands (KBr, cm^{-1}): ν(N^1–H) 3441sb, ν(C–H) 2970w, 2933w, 2812m, 2720w; ν(C=N) + ν(C=C) + δ(N−H) + ν(N=O) 1604m; δ(C−H) 1500m; 1380s, δ(N=O) 1350m; 1279w, 1288w, 1150w, 915w, 529w; ν(C–S) 765s; 650w, 837w. Electronic absorption spectrum (DMSO, λ_{max} /nm, ε / L mol^{-1} cm^{-1}): [10^{-4} M] 419 (2.95 x 10^3), 366 (4.08 x 10^3), 284 (1.03 x 10^4). Fluorescence spectrum: (λ_{max}^{em} = 443 nm; λ_{max}^{ex} = 340 nm). ^{1}H NMR (δ, ppm; $CDCl_3$: DMSO; 9:1): δ = 8.53 (2H, s, C^7H_{bipy}+ $C^{14}H_{bipy}$); 8.31 (H, d, C^2H), 8.17 (2H, d, C^4H+C^6H), 7.92 (3H, m, $C^{10}H_{bipy}$ + $C^{11}H_{bipy}$+ N^1H), 7.67 (2H, d, C^9H_{bipy} + $C^{12}H_{bipy}$), 7.43 (2H, s, C^8H_{bipy} + $C^{13}H_{bipy}$), 6.29 (1H, s, C^5H), 3.30 (2H, m, $N^1(CH_2)$, 1.83 (3H, m, CH_3). ESI mass data: calcd for $C_{20}H_{18}N_6O_3SZn{\cdot}H_2O$, [Zn(3-$NO_2$-stsc-$N^1$HEt)(bipy)+H]$^+$.$H_2O$ **,** m/z = calcd 505.06; obsd. 505.02.

[Zn(3-NO_2-stsc-N^1HPh)(bipy)] (4). Yield, 0.049 g, 79%, mp 221-223°C. Elemental analysis for $C_{24}H_{18}N_6O_3SZn$: calcd C 53.79; H 3.24; N 15.68; S 5.98(%); found: C 53.95; H 3.48; N 15.81; S 6.14%. Main IR Bands (KBr, cm^{-1}): ν(N^1–H) 3408 sb; ν(C–H) 3108w, 3000w; ν(C=N) + ν(C=C) + δ(N−H) 1604s, 1600s; ν(N=O) 1546m; δ(C-H) 1479s 1429s, 1404s; δ(N=O) 1316s; 1279m, 1238m, 1150m, 1100w, 1030w, 966w, 870w, 833m; ν(C–S) 766s; 691m, 658w, 587m, 508m, 420m. Electronic absorption spectrum (DMSO, λ_{max} /nm, ε / L mol^{-1} cm^{-1}): [10^{-4} M] 421 (9.39 x 10^3), 363 (1.58 x 10^4), 282 (1.49 x 10^4). Fluorescence spectrum: (λ_{max}^{em} = 439 nm; λ_{max}^{ex} = 340 nm). ^{1}H NMR (δ, ppm; $CDCl_3$): δ = 8.89 (2H, s, C^7H_{bipy}+ $C^{14}H_{bipy}$), 8.65 (1H, s, C^2H),

8.49 (2H, s ,$C^{10}H_{bipy}$+ $C^{11}H_{bipy}$), 8.20 (2H, d, $C^{9}H_{bipy}$+ $C^{12}H_{bipy}$), 8.05 (2H, s, $C^{4}H$ + $C^{6}H$), 7.91 (3H, s, $C^{8}H_{bipy}$+ $C^{13}H_{bipy}$+ $N^{1}H$), 7.62 (2H, d, *o*-H_{Ph}), 7.39 (2H, m, *m*-H_{Ph}), 6.97 (1H, m, *p*-H_{Ph}), 6.32 (1H, s, $C^{5}H$).

[Zn(3-NO_2-stsc-N^1H_2)(phen)] (**5**).Yield, 0.040 g, 73%, mp 199-201°C. Elemental analysis for $C_{20}H_{14}N_6O_3SZn$: calcd C 49.65; H 2.92; N 17.37; S 6.63(%); found: C 49.69; H 2.74; N 17.63; S 6.87. Main IR bands (KBr, cm^{-1}): ν(N^1–H_2) 3437sb; ν(C–H) 2970w, 2920w, 2812m, 2721w; ν(C=N) + ν(C=C) + δ(N−H) 1600s; ν(N=O) 1521m; δ(C−H) 1474m, 1425w; 1387s, δ(N=O) 1354w; 1233m, 1142w, 1100w, 1087w, 970w; 866w; ν(C–S) 766s; 725s, 642s, 583m, 454w, 425m, 404w. Electronic absorption spectrum (DMSO, λ_{max} /nm, ε / L mol^{-1} cm^{-1}): [10^{-4} M] 411 (5.42 x 10^3), 371 (0.57 x 10^4), 294 (1.09 x10^4), 266 (2.45 x 10^4). Fluorescence spectrum: (λ_{max}^{em} = 443 nm; λ_{max}^{ex} = 340 nm). ^{1}H NMR (δ, ppm; $CDCl_3$: DMSO; 9:1): δ = 8.93 (2H, s, $C^{7}H_{phen}$ + $C^{14}H_{phen}$); 8.64 (1H, s, $C^{2}H$), 8.54 (2H, s, $C^{9}H_{phen}$ + $C^{12}H_{phen}$), 7.99 (2H, d, $C^{4}H$ + $C^{6}H$), 7.86 (2H, s, $C^{10}H_{phen}$ + $C^{11}H_{phen}$), 7.65 (2H, m, $C^{8}H_{phen}$ + $C^{13}H_{phen}$), 7.59 (1H, s, $N^{1}H$), 6.41(1H, s, $C^{5}H$). ESI mass data: calcd for $C_{20}H_{14}N_6O_3SZn$, [Zn(3-NO_2-stsc-N^1H_2)(phen)+H]$^+$ **,** m/z = calcd 483.02, obsd. 482.99.

[Zn(3-NO_2-stsc-N^1HMe)(phen)] (**6**). Yield, 0.041 g, 73%, mp 208-210°C. Elemental analysis for $C_{21}H_{16}N_6O_3SZn$: calcd C 50.66; H 3.24; N 16.88; S 6.44 %; found: C 50.84; H 3.38; N 16.62; S 6.08%. Main IR bands (KBr, cm^{-1}): ν(N^1–H) 3420sb; ν(C–H) 2970w, 2937w, 2816m, 2725m; ν(C=N) + ν(C=C) + δ(N-H) 1608s; ν(N=O) 1533m; δ(C-H) 1478s, 1500s, 1487s, 1425w; 1387s; δ(N=O) 1350s; 1283w, 1233m, 1170w, 970w; ν(C–S) 746s; 725s, 646s, 587w, 538w, 462w, 429m, 408w. Electronic absorption spectrum (DMSO, λ_{max} /nm, ε / L mol^{-1} cm^{-1}): [10^{-4} M] 425 (6.58 x 10^3), 363 (1.07 x 10^4), 293 (1.13 x 10^4), 263 (3.11 x10^4). Fluorescence spectrum: (λ_{max}^{em} = 473 nm; λ_{max}^{ex} = 340 nm). ^{1}H NMR (δ, ppm; $CDCl_3$: DMSO; 9:1): δ = 9.00 (2H, s,

C^7H_{phen} + $C^{14}H_{phen}$), 8.61 (2H, s, C^9H_{phen} + $C^{12}H_{phen}$), 8.01 (2H, $C^{10}H_{phen}$ + $C^{11}H_{phen}$), 7.94 (1H, d, C^2H), 7.54 (2H, m, C^4H+C^6H), 7.51(1H, m, N^1H), 7.38 (2H, m, C^8H_{phen} + $C^{13}H_{phen}$), 6.41 (1H, s, C^5H), 3.00 (3H, d, CH_3N^1)).

[Zn(3-NO_2-stsc-N^1HEt)(phen)] (7). Yield, 0.045 g, 77%, mp 195-197°C. Elemental analysis for $C_{22}H_{18}N_6O_3SZn$: calcd C 51.62; H 3.54; N 16.42; S 6.26%; found : C 51.72; H 3.43; N 16.21; S 6.07%. Main IR bands (KBr, cm^{-1}): ν(N^1–H) 3441br; ν(C–H) 2967w, 2929w, 2816w, 2725w; ν(C=N) + ν(C=C) + δ(N−H) + ν(N=O) 1600sb; 1513s; δ(C-H) 1450w, 1423s; 1387s; δ(N=O) 1354s; 1291w, 1233w, 1154w, 1100w, 854w; ν(C–S) 775s; 729w, 600w, 416w; UV-vis data DMSO, λ_{max} /nm, ε / L mol^{-1} cm^{-1}: [10^{-4}M] 428 (6.63 x 10^3), 361 (1.01 x 10^4), 295 (1.01 x 10^4), 258 (2.86 x 10^4). Fluorescence data : (λ_{max}^{em} = 444 nm; λ_{max}^{ex} = 340 nm). ^{1}H NMR (δ, ppm; $CDCl_3$: DMSO; 9 : 1): δ = 8.75 (2H, s, C^7H_{phen} + $C^{14}H_{phen}$), 8.39 (3H, d, C^2H+ C^9H_{phen} + $C^{12}H_{phen}$), 7.82 (2H, m, C^4H+C^6H), 7.71 (3H, m, $C^{10}H_{phen}$ + $C^{11}H_{phen}$ + N^1H), 7.35 (2H, s, C^8H_{phen} + $C^{13}H_{phen}$), 6.15 (1H, s, C^5H), 3.28 (2H, m, $N^1(CH_2)$, 1.84 (3H, m, CH_3).

[Zn(3-NO_2-stsc-N^1HPh)(phen)] (8). Yield, 0.050 g, 79%, mp 225-227°C. Elemental analysis for $C_{26}H_{18}N_6O_3SZn$:calcd C 50.29; H 3.13; N 13.03; S 4.97%; found: C 50.21; H 3.26; N 13.19; S 4.88 %. Main IR bands (KBr, cm^{-1}): ν(N^1–H) 3400sb; ν(C–H) 2808w, 2721w; ν(C=N) + ν(C=C) + δ(N−H) + 1608s, 1546s; ν(N=O) 1546m; δ(C−H) 1479m, 1429s; 1383s; δ(N=O) 1350s; 1279m, 1233m, 1196w, 1138w, 970s, 870m; ν(C–S) 766s; 742s, 705s, 700w, 642w, 596w, 496w, 421w. Electronic absorption spectrum (DMSO, λ_{max} /nm, ε / L mol^{-1} cm^{-1}): [10^{-4} M] 427 (8.99 x 10^3), 364 (1.59 x 10^4), 294 (1.03 x a0^4), 266 (3.91 x 10^4). Fluorescence spectrum: (λ_{max}^{em} = 438 nm; λ^{ex} = 340 nm). ^{1}H NMR (δ, ppm; $CDCl_3$: DMSO, 9:1): δ = 8.80

(2H, s, C^7H_{phen} + $C^{14}H_{phen}$), 8.42 (1H, s, C^2H), 8.36 (2H, d, C^9H_{phen} + $C^{12}H_{phen}$), 8.32 (2H, d, $C^{10}H_{phen}$ + $C^{11}H_{phen}$), 8.01 (2H, s, C^4H + C^6H), 7.99 (1H, m, N^1 H), 7.89 (2H, m, C^8H_{phen} + $C^{13}H_{phen}$), 7.76 (2H, d, o-H_{Ph}), 7.40 (2H, m, m-H_{Ph}), 7.13 (1H, m, p-H_{Ph}), 6.24 (1H, d, C^5H). ESI mass data: calcd for $C_{26}H_{18}N_6O_3SZn$, $[Zn(3\text{-}NO_2\text{-}stsc\text{-}N^1HEt)(phen) + H]^+$, m/z = calcd 559.05; obsd. 559.01.

X –ray crystallography

A single crystal of a complex was mounted on a polymer loop and the data were measured using a Rigaku Oxford Diffraction four-circle diffractometer equipped with the graphite monochromated Cu-Kα (λ = 1.54184 Å; **4, 5**) and Mo-Kα (λ = 0.71073 Å; **7, 8**) radiation sources. The crystal data were collected at 173(2) K (**4, 5, 7** and **8)** and processed with CrysAlisPro (data collection, data reduction and cell refinement). The structures were solved by the direct methods using the program OLEX2 1.2 and refined by the full-matrix least-squares technique based on F^2 using SHELXT.[37-40] Molecular graphics were produced with Olex2. All the non-hydrogen atoms were refined anisotropically, while all hydrogen atoms were fixed geometrically with their U_{iso} values 1.2 times except for the ones attached to the imidazole nitrogens / aromatic rings.

Antimicrobial studies

The reference strains of bacteria and yeasts were obtained from Microbial Type Culture Collection (MTCC), Institute of Microbial Technology (IMTECH), Chandigarh, India and the clinical isolate methicillin resistant *Staphylococcus aureus* (MRSA) was obtained from the Post graduate Institute of Medical Education and Research, (PGIMER), Chandigarh, India. Reference

strains included methicillin resistant *Staphylococcus aureus* (MRSA - a clinical isolate of resistant bacteria), Gram positive bacteria, viz. *Staphylococcus aureus* (MTCC740), Gram negative bacteria, *Klebsiella pneumoniae* 1 (MTCC109), *Salmonella typhimurium* 2 (MTCC1251) and a yeast, *Candida albicans* (MTCC227). The minimum inhibitory concentration (MIC) and zone of inhibition of the selected compounds was worked out by the agar dilution method.[41] The colorimetric measurements of 3-[(4, 5-dimethylthiazol-2-yl)-2, 5-diphenyl] tetrazolium bromide (MTT) cytotoxicity assay is based on the capacity of mitochondrial succinate dehydrogenase enzymes in living cells (sheep blood used) to reduce the yellow water soluble substrate (MTT) into an insoluble purple colored formazan product that are dissolved in DMSO.[42] (The detailed procedures of the study are similar to those reported in our previous work).[25]

Conflict of Interest

There are no conflicts of interest to declare.

Acknowledgments

Financial assistance from the University Grant Commission (BSR, UGC); New Delhi is gratefully acknowledged. Jerry P. Jasinski acknowledges the NSF–MRI program (Grant No. CHE-1039027) for funds to purchase the X-ray diffractometer. Tarlok Singh Lobana thanks the Guru Nanak Dev University for Honorary Professor position.

References

1 C. A. Kontogiorgis, P. Papaioannou, D. J. Hadjipavlou-Litina, *Curr. Med. Chem.* 2005, **12**, 339-355.

2 A. Scozzafava, C.J. Supuran, *J. Med. Chem.* 2000, **43**, 3677-3687.

3 Y. –H. Chiu, G. J. Gabriel, J.W. Canary, *Inorg. Chem.* 2005, **44**, 40-44.

4 T. A. Miller, D.J. Witter, S.J. Belvedere, *J. Med. Chem.* 2003, **46**, 5097-5116.

5 T. Suzuki, A. Matsuura, A. Kouketsu, H. Nakagawa, N. Miyata, *Bioorg. Med. Chem. Lett.* 2005, **15**, 331-335.

6 E. Kimura, T. Koike, *Adv. Inorg. Chem.* 1997, **44**, 229-261.

7 S.V. Ghuge, M.C. Zarkar, S.R. Deshmukh, S.K. Choubey, *J. Adv. Bioinf. Appl. Res.* 2010, **1**, 17-26.

8 A. K. Patick, K.E. Potts, *Clin. Microbiol. Rev.* 1998, **11**, 614-627.

9 T. S. Lobana, R. Sharma, G. Bawa, S. Khanna, *Coord. Chem. Rev.* 2009, **253,** 977-1055.

10 T. S. Lobana, *RSC Adv.* 2015, **5**, 37231-37274.

11 S. Datta, D. K. Seth, S. Gangopadhyay, P. Karmakar, and S. Bhattacharya, *Inorg. Chim.*

Acta 2012, **392,** 118-130.

12 R. Saswati, C. Dinda, S. Schmiesing, E. Sinn, Y. P. Patil, M. Nethaji, H. S. Evans, and R. Acharya, Polyhedron 2013, **50,** 354-363.

13 S. I. Pascu, P. A. Waghorn, B. W. C. Kennedy, R. Arrowsmith, S. R. Bayly, J. R. Dilworth, M. Christlieb, R. M. Tyrrell, J. Zhong, R. M. Kowalczyk, D. Collison, P. K. Aley, G.C. Churchill, and F. I. Aigbirhio, *Chem.- Asian J.* 2010, **5,** 506-519.

14 M. Christlieb, A. R. Cowley, J. R. Dilworth, P. S. Donnelly, B. M. Paterson, H.S.R. Struthers, and J. M. White, *Dalton Trans.* 2007, 327-331.

15 K. Alomar, A. Landreau, M. Kempf, M. A. Khan, M. Allain and G. Bouet, J. *Inorg. Biochem.* 2010, **104**, 397–404.

16 N. C. Kasuga, K. Sekino, M. Ishikawa, A. Honda, M. Yokoyama, S. Nakano, N. Shimada, C. Koumo, K. Nomiya, *J. Inorg. Biochem.* 2003, **96**, 298–310.

17 M. C. Rodríguez-Argüelles, P. T. Touceda, R. Cao, A. M. García-Deibe, P. Pelagatt, C. Pelizzi, F. Zani, *J. Inorg. Biochem.* 2009, **103**, 35–42.

18 E. Bermejo, R. Carballo, A. Castineiras, R. Dominguez, C. Maichle-Mossmer, J. Strahle, D.X. West, *Polyhedron* 1999, **18**, 3695-3702.

19 Y. Li, Z. –Y. Yang, J. –Cai Wu, *Eur. J. Med. Chem.* 2010, **45**, 5692-5701.

20 M. C. Rodriguez-Arguelles, M.B. Ferrari, G.G. Fava, C. Pelizzi, P. Tarasconi, R. Albertini, P. P. Dall'Aglio, P. Lunghi, S. Pinelli, *J. Inorg. Biochem.* 1995, **58**, 157-175.

21 M. B. Ferrari, F. Bisceglie, G. Pelosi, P. Tarasconi, R. Albertini, S. Pinelli, *J. Inorg. Biochem.* 2001, **87**, 137-147.

22 D. K.-Demertzi, P. N. Yadav, J. Wiecek, S. Skoulika, T.Varadinova, M. A. Demertzis, *J. Inorg. Biochem.* 2006, **100**, 1558–1567.

23 U. El-ayaan, J. *Coord. Chem.* 2012, **65**, 629–642.

24 Y. Li, Z.-Y. Yang, J.-C. Wu, *Eur. J. Med. Chem.* 2010, **45**, 5692-5701.

25 S. Indoria, T. S. Lobana, Henna Sood, D. S. Arora, G. Hundal and J. P. Jasinski, *New J. Chem.* 2016, **40**, 3642-3653.

26 M. Kaushal, S. Indoria, T.S. Lobana, H. Sood, D.S. Arora, G. Hundal,V.A. Smolinski, M. Kaur and J.P. Jasinski, *Polyhedron*, 2018, **148**, 9-21.

27 A. B. P. Lever, Inorganic electronic spectroscopy (second Edition) Elsevier, New York, 1984, 636.

28 A. W. Addison, T. N. Rao, J. Reedijk, J. Rijn, and G. C. Verschoor, *J. Chem. Soc. Dalton Trans*. 1984, 1349-1356.

29 J.E. Huheey, E.A. Keiter and R.L. Keiter, *Inorganic Chemistry: Principles of Structure and Reactivity,* Harper Collins College Publishers, New York, 4th edn, (1993).

30 J.K. Aulakh, T.S. Lobana, Henna Sood, D.S. Arora , Isabel Garcia-Santos, G. Hundal, M. Kaur, V.A. Smolenski, and Jerry P. Jasinski, *Dalton Trans.*, 2017, **46**, 1324–1339; ; ibid.

Dalton Trans. 2017, **46**, 15138.

31 T. S. Lobana, S. Indoria, A. K. Jassal, H. Kaur, D. S. Arora and J. P. Jasinski, *Eur. J. Med. Chem*., 2014, **76**, 145–154.

32 T. S. Lobana, Shikha Indoria, H. Kaur, D. S. Arora, A. K. Jassala and J. P. Jasinski, *RSC Adv.*, 2015, **5**, 14916–14936

33 G. Ciapetti, E. Cenni, L. Pratelli and A. Pizzoferrato, *Biomaterials*, 1993, **14,** 359–364.

34 F.E. Andersen, C.J. Duca, J.V. Scudi, *J. Am. Chem. Soc.* 1951, **73**, 4967 - 4968.

35 M.B. Ferrari, S. Capacchi, G. Pelosi, G. Reffo, P. Tarasconi, R. Albertini, S. Pinelli, P. Lunghi, *Inorg. Chim. Acta* 1999, **286**, 134 - 141.

36 E. Pahontu, V. Fala, A. Gulea, D. Poirier, V. Tapcov, T. Rosu, Molecules 2013, **18**, 8812 - 8836.

37 G.M. Sheldrick, *Acta Crystallogr. Sect. A* 2008, **64**, 112-122.

38 L. Palatinus, S.J. Prathapa and S. van. Smaalen, *J. Appl. Crystallogr.* 2012, **45**, 575-580.

39 L. Palatinus and A. van der Lee, *J. Appl. Crystallogr.* 2008, **41**, 975-984.

40 O.V. Dolomanov, L.J. Bourhis, R.J. Gildea, J.A.K. Howard, and H. Puschmann, *J. Appl. Crystallogr.* 2009, **42**, 339-341.

41 G. J. Kaur and D. S. Arora, *BMC Complement Altern. Med*. 2009, 09 -30.

42 J. G. Onsare and D. S. Arora, *J. Appl. Microbiol.,* 2014, **118**, 313–325.