Synthesis, crystal structure, antibacterial activity and theoretical studies on a novel mononuclear cobalt(II) complex based on 2,4,6-tris(2-pyridyl)-1,3,5-triazine ligand

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HIGHLIGHTS

• A new Co(II) complex with formula [Co(tptz)(CH3OH)Cl2] CH3OH 0.5H2O has been prepared.
• Using tptz = 2,4,6-tris(2-pyridyl)-1,3,5-triazine as ligand.
• The antibacterial activity of Co(II), Ni(II), Cu(II), Mn(II) and Rh(III) with tptz have been evaluated.
• The DFT studies consistent with experimental results.

GRAPHICAL ABSTRACT

The crystal structure of Co(II) with formula [Co(tptz)(CH3OH)Cl2] CH3OH 0.5H2O, (tptz = 2,4,6-tris(2-pyridyl)-1,3,5-triazine) was determined by single-crystal X-ray diffraction. The DFT studies consistent with experimental results. Then, the in vitro antibacterial activity of ligand and tptz complexes of Co(II), Ni(II), Cu(II), Mn(II) and Rh(III) have been evaluated.

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ABSTRACT

A cobalt complex was prepared from CoCl2 6H2O and 2,4,6-tris(2-pyridyl)-1,3,5-triazine (tptz) in methanol and designated as [Co(tptz)(CH3OH)Cl2] CH3OH 0.5H2O (1). It was characterized by several techniques including TGA analysis and FT-IR, UV–Vis and 1H NMR spectral studies. The crystal structure of 1 was determined by single-crystal X-ray diffraction. The Co(II) metal center in 1 is six coordinated with a distorted octahedral geometry. The tptz ligand is tridentate and coordinates to the cobalt through coplanar nitrogen atoms from the triazine and two pyridyl rings. Two chloride anions and a methanol molecule complete the inner coordination sphere of the metal ion. The optimized geometrical parameters obtained by DFT calculation are in good agreement with single XRD data. The in vitro antibacterial activity of various tptz complexes of Co(II), Ni(II), Cu(II), Mn(II) and Rh(III) were evaluated against Gram-positive (Bacillus subtilis, Staphylococcus aureus and Gram-negative (Escherichia coli and Pseudomonas aeruginosa) bacteria. Whereas all complexes exhibited good activity in comparison to standard antibacterial drugs, the inhibitory effects of complexes were found to be more than that of the parent ligand. Overall, the obtained results strongly suggest that the cobalt(II) complex is a suitable candidate for counteracting antibiotic resistant microorganisms.

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Introduction

Coordination compounds exhibit different characteristic properties which depend strongly on the nature of the ligands and the coordination behavior of the metal atoms. Synthesis and Evaluation of the biological activity of metal complexes incorporating biologically active ligands is subject of much current interest [1–3]. The use of transition metal complexes as therapeutic compounds has become more and more pronounced. These complexes offer a great diversity in their actions. However, the development of transition metal complexes as drugs is not an easy task; considerable effort is required to obtain a compound of interest. In general, the ultimate goal is to offer appropriate and efficient antibacterial agents to counteract antibiotic resistant microorganisms [4]. Potential metal–antibiotic drugs were focused in line with application of bio-coordination chemistry which appears to be crucial for improving the design of compounds to reduce toxic side effects and understand their mechanisms of action. This serves as a light cast for chemists interested in developing greener design of drugs.

For centuries, people have used copper and cobalt ions to inhibit the growth of harmful microbes [5]. Cobalt is a physiologically relevant metal that plays an important role in many biological processes and exhibits considerable biochemical action as an essential trace metal among all other transition metal ions [6]. As such, synthesis of new cobalt complex drugs have found extensive applications in various fields of human interest such as antimicrobial [7], antiviral [8], antifungal [8], antioxidant activity [9], anti-inflammatory [10], transport protein transferrin (Tf) [11], antymicrobacterial [12], antischismic [13], antiparasitic [14], antithrombolytic [14], enzymatic therapeutic agents [14] or even antitumor agents [11,15] due to their biological role and synergistic activity with other drugs. Therefore, study and development of new cobalt complexes could be helpful in design and production of antibacterial agents to counteract antibiotic resistant microorganisms [16].

Derivatives of 1,3,5-triazine have received much attention as they have shown promising potential as antitumor and antibacterial agents [1,17]. 2,4,6-Tri(2-pyridyl)-triazine (tptz, see Scheme 1 in supplementary) is of interest as it is a bulky aromatic compound which has three 2-pyridyl rings bonded to the central 1,3,5-triazine platform [18]. The planar structure of tptz along with its large π system also makes it a suitable candidate for further chemical modifications. Tptz has potential in pharmacologically active coordination compounds and has been shown to be a useful ligand in coordination chemistry [19].

The aim of the present study was the synthesis and crystal structure determination of a cobalt(II) complex with the tptz ligand followed by further investigation of its properties by spectrophotometric and computational studies. The antibacterial potential of the newly synthesized cobalt complex together with a number of other tptz complexes of biologically important metal ions, such as nickel, copper, manganese, rhodium complexes with tptz were also studied [20–23]. These investigations screened the complexes for their antibacterial activities against both Gram positive and Gram negative bacteria [20–24].

Materials and methods

Reagents and techniques

All materials were of commercial reagent grade and were used without further purification. Elemental analyses were performed on a Heraeus CHN-O-Rapid elemental analyzer. FTIR spectra of the samples were recorded on a FTIR JASCO 460 spectrophotometer using KBr pellet in the range of 4000–400 cm\textsuperscript{-1} and electronic spectra were obtained using a JASCO 7580 spectrophotometer. \textsuperscript{1}H NMR spectra were recorded on a Bruker DRX-300 MHz, at ambient temperature in DMSO-\textit{d}_6. Thermal analysis measurements, TGA, were recorded on a Shimadzu model 50 instrument using 20 mg of [Co(tptz)(CH\textsubscript{3}OH)Cl\textsubscript{2}] CH\textsubscript{3}OH 0.5H\textsubscript{2}O (I). The oxygen flow rate was 10 cm\textsuperscript{3} min\textsuperscript{-1}.

Synthesis of [Co(tptz)](CH\textsubscript{3}OH)Cl\textsubscript{2}] CH\textsubscript{3}OH 0.5H\textsubscript{2}O (I)

A methanolic solution of CoCl\textsubscript{2}·6H\textsubscript{2}O (237 mg, 1 mmol in 15 mL ethanol) was added dropwise with stirring to a solution of tptz (312 mg, 1 mmol in 15 mL methanol) in a molar ratio of 1:1. The resulting mixture was further stirred for 2 h at room temperature and then heated at reflux on a water bath for 3 h. The brown solid product was collected by suction filtration on a fine frit, washed with methanol and diethyl ether, and then air dried. For further purification, the brown microcrystalline precipitate was recrystallized by diffusion of ether into the solution of the complex in methanol. After 4 days at room temperature, brown crystals were obtained (411 mg, 80% yield), Decompr. Point. 380 °C. Anal. Calc. for Co\textsubscript{2}H\textsubscript{2}Cl\textsubscript{2}CoN\textsubscript{2}O\textsubscript{2s}0 (MW = 515.26): C. 46.57; H. 4.07; N. 16.30. Found: C. 46.57; H. 3.95; N. 16.56%. IR (KBr pellet, cm\textsuperscript{-1}): 3406 (O–H), 3062 (–H aromatic and aliphatic), 1578, 1562 and 1530 (C=N), 1375 (C=C), UV–Vis (MeOH, \textit{λ}_{max}/nm): 400, 290, 250, 211.

Preparation of other complexes with tptz

Other transition metal complexes of tptz, [Ni(tptz)(CH\textsubscript{3}OH)Cl\textsubscript{2}] (2), [Cu(tptz)Cl\textsubscript{2}]2H\textsubscript{2}O (3), [MnCl\textsubscript{2}(tptz)] (4) and [Rh(tptz)Cl\textsubscript{3}]·2H\textsubscript{2}O (5) were prepared using the previously described procedures [20–23].

In vitro antibacterial assay

The in vitro antibacterial activity was investigated against two Gram-positive (Bacillus subtilis ATCC 6633 and Staphylococcus aureus ATCC 6538) and two Gram-negative (Escherichia coli ATCC 35218 and Pseudomonas aeruginosa ATCC 27853) bacteria. In order to compare the results, nalidixic acid (30 mg/disk) and vancomycin (30 mg/disk) were used as standard antibacterial drugs. The determination was carried out by the paper-disk diffusion method [25]. The compounds were dissolved in DMSO at 25 mg/ml concentration. Muller Hinton broth was used to prepare the basal media for the bioassay of the organisms. A lawn culture from a 0.5 MacFarland suspension of each strain was prepared on Muller Hinton agar. Blank paper disks (6.4 mm diameter) were saturated with a solution of the test compounds and placed on the surface of the agar plates. DMSO was poured on to one paper disk only as a control. The plates were incubated at 37 °C for 24 h. The inhibition zone diameters around each disk were measured in mm.

X-ray crystallography

Brown crystals of [Co(tptz)(CH\textsubscript{3}OH)Cl\textsubscript{2}] CH\textsubscript{3}OH 0.5H\textsubscript{2}O (I) were grown by diffusion of ether into a methanolic solution of the complex. The X-ray measurements were carried out on a Bruker APEXII Kappa CCD single crystal diffractometer equipped with a graphite monochromator. Mo Kα radiation (\textit{λ} = 0.71073 Å) was used for the collection, which was controlled by APEX2 [26], with data collected at 94(2) K. The data were corrected for Lorentz and polarization effects using SAINT [26] and numerical absorption corrections were applied using SADABS [26]. Crystals of this material were very weakly diffraacting and, despite experimenting with a variety of samples, no significant data could be obtained beyond θ = 19.26°. It is possible that this may be due to partial loss of the methanol or water solvent molecules from the crystal. The paucity
of data results in a low data: parameter ratio for the structure but the overall identity of the complex is clearly revealed as the structure solved and refined acceptably. The data quality is clearly reflected in the standard uncertainties in the metrical data.

The structure was solved by direct methods using SIR2004 [27] and refined using full-matrix least-squares procedures with SHELXL-97 & TITAN2000 [28,29]. H atoms were positioned geometrically (C–H = 0.95 or 0.98 Å) and refined as riding, with Uiso(H) = 1.2Ueq(C) or 1.5Ueq (methyl C), allowing for free rotation of the methyl groups. The difference map following the location of all non-hydrogen atoms of the Co complex revealed a number of additional high peaks. These were representative as a molecule of methanol solvent on a general position with the O atom and a partial occupancy water molecule that was disordered over two positions. Refinement of the occupancies converged at an occupancy ratio approximately 0.3:0.2 and the occupancies of the O atoms were fixed at these values in the final refinement cycle. It was not possible to successfully model the H atom positions for the disordered water molecule.

Molecular plots and packing diagrams were produced using Mercury [30] and other metrical data was produced using PLATON [31].

**Computational details**

All calculations were performed using the Gaussian 98 [32] software. The geometry optimization of ligand and all the complexes were performed employing a hybrid Hartree–Fock-density functional scheme, the adiabatic connection method-Becke three-parameter with Lee–Yang–Parr (B3LYP) functional of density functional theory (DFT) using the Hartree–Fock-density functional scheme [33,34] with the three standard 6-31G*, 6-311G** and 6-311+G** basis sets. Restricted formalism was applied to all close-shell systems. For the [Co(tptz)(CH3OH)Cl2] complex, calculations were performed with the unrestricted formalism (UB3LYP). Full optimizations were performed without any symmetry constrains. Harmonic vibrational frequencies were computed to confirm that an optimized geometry correctly corresponds to a local minimum that has only real frequencies. In addition, the thermodynamic properties of all compounds were obtained from frequency calculations at 298.15 K and 1.0 atmosphere pressure. All reported enthalpies were zero-point (ZPE) corrected with unscaled frequencies. The solvent effects on the conformational equilibrium and contribution to the total enthalpies were investigated with using polarized continuum (overlapping spheres) model (PCM) of Tomasi and coworkers [35] at the B3LYP/6-31G* level. Solvation calculations were carried out for methanol (ε = 32.7) with the geometries optimization for this solvent. The POP = NBO key-word is used for a full natural bond orbital analysis of spin density and atomic charge assignments [36].

**Results and discussions**

**Characterization of [Co(tptz)(CH3OH)Cl2]CH3OH 0.5H2O (1)**

The complex 1 was prepared in high yield with high purity by combination of a methanolic solution of CoCl2·6H2O and tptz in a stoichiometric ratio of 1:1 at room temperature. It was readily recrystallized by diffusion of ether into a methanolic solution of the complex. The complex was characterized by FT-IR, UV–Vis, and 1H NMR and mass spectrometry, TGA, elemental analysis and X-ray crystallography. The elemental analysis results for complex 1 were found to be entirely consistent with its composition as determined by X-ray crystallography.

**Crystal structure of [Co(tptz)(CH3OH)Cl2]CH3OH 0.5H2O (1)**

A perspective view of the molecule [Co(tptz)(CH3OH)Cl2]CH3OH 0.5H2O (1) with the atom numbering scheme is shown in Fig. 1. Details of the crystal, the data collection, selected bond distances and angles are given in Tables 1, S1 and 2 respectively. The asymmetric unit of the structure comprises the [Co(tptz) (CH3OH)Cl2] complex, together with a methanol molecule and a water molecule that refined to half occupancy and is disordered over two positions.

In the structure of complex species (Fig. 1), a single Co(II) metal center binds to the tptz ligand in a tridentate fashion through a single N atom from the central triazine ring and N atoms from two of the three pyridyl rings. Two chloride ions and the O atom of a methanol molecule complete the octahedral coordination sphere of the Co(II) cation. One chloride ion, Cl1, is trans to the N1 atom of the triazine and the second Cl− anion mutually trans. As observed in similar complexes [37], the tptz ligand is close to planar with an rms deviation of 0.086 Å from the best fit plane through all 24 ligand atoms. The ligand plane also lies close to the equatorial plane of the complex defined by the C11, N1, N4 and N5 donor atoms with a dihedral angle of 5.14(7)° between them. Within the tptz ligand itself the central triazine ring submends angles of 4.4 (2)° and 6.4(2)° to the coordinated N4 and N5 pyridyl rings respectively and 3.9(2)° to the pendant N6 ring. The N(1)−Co−N(4) and N(1)−Co−N(5) bite angles 73.98(14) and 74.30(14) are significantly smaller than the ideal value of 90° because of the constraint imposed by the five membered chelate rings. However these compare well with the average value of 74.4(3)° found for the N1(C1)tpzt complexes found in the Cambridge Structural Database [38](Version 5.35 November 2013 with three updates). Other similarities to previously reported Co(II) tpzt complexes include the short Co1−N1 bond, 2.079(4) Å compared to Co−N4, 2.211 (4) and Co−N5, 2.218 (4) involving the two pyridyl rings. Furthermore, the Co1−C1 bond at 2.3124(13) Å is significantly shorter than Co1−C2, a similar bond length variation was found in the only other CoCl2 complex of the tpzt ligand to be reported in the literature [39]. In the crystal structure, an extensive array of classical O−H⋯N and O−H⋯O hydrogen bonds involving both the ligated and solvate methanol molecules and the solvent water combine with non-classical C–H⋯Cl and C–H⋯O contacts to form a three dimensional network with layers of complex molecules parallel to the ab diagonal, Fig. 2 and Table 3. Offset π−π stacking interactions are also found with centroid– centroid distances Cg3⋯Cg4 = 3.672(3) Å, Cg2⋯Cg6 = 3.613(3) Å and Cg5⋯Cg5 = 3.90(3) Å (Cg3, Cg4 and Cg5 are the centroids of the N1, C1, N2, C13, N3, C7, N4, C2, C3, C4, C5, C6 and N5, C8, C9, C10, C11, C12 rings respectively; symmetry operations v = 1 − x, 1 − y, 1 − z; vi = 2 − x, 1 − y, 1 − z; vii = 1 − x, − y, − z).

**UV–Vis and IR**

The electronic absorption spectrum of [Co(tptz)(CH3OH)Cl2]CH3OH 0.5H2O (1) in methanol was recorded in the range 200–800 nm (Fig. 3) and the data are presented in the experimental section. In the UV region, the complex exhibits three intense absorption bands at 211 nm, 290 nm and 250 nm. Compared to the absorption spectrum of free tptz, it provides an evidence that these sharp bands (shifted nearly 10 nm compared to those in the free ligand) are ligand-centered (LC) due to π → π∗ or n → π∗ transitions of the metal-bound tptz chromophores, which are only somewhat perturbed by complexation [40]. Furthermore, the complex shows one broad band with very low absorptivity centered at 400 nm. This is broader and less intense and can be assigned to a d–d transition from the 3d7 cobalt(II) metal center [41].
The FT-IR spectra of free tptz and compound 1 are shown in Fig. 4(a and b), respectively. The FT-IR spectrum of free tptz displays three bands at 1617, 1524 and 1468 cm\(^{-1}\) due to the \(\nu(C=N)\) stretching mode of the triazine ring, and the \(\nu(C=C)\) and \(\nu(C=N)\) modes of the pyridyl rings, respectively [20]. The intense band at 1367 cm\(^{-1}\) is assigned to the stretching vibrations of both

![Fig. 1. (a) ORTEP view of the mononuclear cobalt complex [Co(tptz)(CH\(_3\)OH)Cl\(_2\)]CH\(_3\)OH.0.5H\(_2\)O (1).](image)
the triazine and pyridine rings of tptz. The FT-IR spectrum of the complex \([\text{Co(tptz)}(\text{CH}_3\text{OH})\text{Cl}_2]\) \(\text{CH}_3\text{OH}0.5\text{H}_2\text{O}\) is very similar to that of the free tptz ligand. The three vibrations for the \(\nu(\text{C}=\text{N})\) stretching modes of the triazine, and the \(\nu(\text{C}=\text{N})\) and \(\nu(\text{C}=-\text{C})\) bands of the pyridyl rings of the coordinated tptz ligand appear at 1483, 1530 and 1558 cm\(^{-1}\). As compared with the free ligand, these bands exhibit slight shifts to lower energy together with a degree of splitting, characteristic of complex formation and the involvement of triazine and pyridyl nitrogens in coordination to the metal ion [39,42]. The vibration at 774 cm\(^{-1}\) is slightly shifted compared to that of the free ligand. The band at 3406 cm\(^{-1}\) is assigned to the \(\nu(\text{C}\equiv\text{N})\) stretching modes of the triazine, and the \(\nu(\text{C}\equiv\text{N})\) band of the coordinated ring (C). The tptz ligand chemical shifts can be described to different coordinated pyridyl rings (A) and (B) and non-coordinated ring (C). The tptz ligand chemical shifts can be described either by contact or pseudo-contact shifts. The first process is a result of unpaired electron spin density being transferred from the paramagnetic Co(II) ion towards the tptz ligand and depends on the magnitude of ligand character in the half-filled molecular orbital. Different protons are subjected to different magnitudes of contact shift due to the delocalization of spin density being unevenly distributed. On the other hand, the pseudo-contact shift arises from the through space interaction between electron and nuclear magnetic moments. The magnitude and sign of the pseudo-contact shift depend on the distance, relative geometry (angles) of each proton from the paramagnetic Co(II) center, and the anisotropy of the magnetic susceptibility \((\chi)\). This shift usually has a negative sign and shifts the proton chemical shifts upfield. Depending on their positions, each particular proton is subjected to a combination of these two types of shifts which can be seen in the chemical shift range of the paramagnetic NMR spectrum. Recall that assignment of the paramagnetic NMR spectra of Co(II) complexes has always been a difficult task as the chemical shift values cannot be easily predicted. The isotropic shift which is the sum of the two contributions and can be opposite in signs is determined by many factors. For instance, the ortho hydrogen atoms \((\text{Ha})\) of the tptz ligand pyridine rings which are nearest to the paramagnetic Co(II) center, are again most affected by the contact shift and are shifted downfield. On the other hand, the meta and para hydrogen atoms are shifted upfield largely because of the pseudo-contact shift [43]. The contribution of contact shift for these protons is small because of their distance from Co(II).

The measurement of the effective magnetic moment of the complex in solution was determined by the Evans method at room temperature NMR spectroscopy [44]. In this method, a solution of

![Fig. 3. Electronic spectra of \([\text{Co(tptz)}(\text{CH}_3\text{OH})\text{Cl}_2]\) \(\text{CH}_3\text{OH}0.5\text{H}_2\text{O}\) in methanol.](image)

![Fig. 4. FT-IR spectra of (a)TPTZ ligand and (b) \([\text{Co(tptz)}(\text{CH}_3\text{OH})\text{Cl}_2]\) \(\text{CH}_3\text{OH}0.5\text{H}_2\text{O}\).](image)

![Fig. 2. The crystal packing of \([\text{Co(tptz)}(\text{CH}_3\text{OH})\text{Cl}_2]\) \(\text{CH}_3\text{OH}0.5\text{H}_2\text{O}\).](image)

### Table 3

| Hydrogen bond distances (Å), angles (°) for \([\text{Co(tptz)}(\text{CH}_3\text{OH})\text{Cl}_2]\) \(\text{CH}_3\text{OH}0.5\text{H}_2\text{O}\) (1). | d(D–H) | d(D–A) | d(H–A) | \(\chi(DHA)\) |
|---|---|---|---|---|
| C(9)–H(9) · O(1S) | 0.95 | 2.72 | 3.57(8) | 150 |
| C(10)–H(10) · O(25A) | 0.95 | 2.67 | 3.30(9) | 125 |
| C(17)–H(17) · Cl(2)v | 0.95 | 2.71 | 3.53(5) | 145 |
| C(11)–H(11) · Cl(1)xii | 0.95 | 2.91 | 3.47(5) | 120 |
| C(5)–H(5) · Cl(1)xvi | 0.95 | 2.88 | 3.79(5) | 162 |
| C(10)–H(10) · Cl(2)xvii | 0.95 | 2.70 | 3.56(5) | 152 |
| C(3)–H(3) · O(1S) | 0.95 | 2.66 | 3.53(5) | 152 |
| O(1)–H(10) · N(2)ii | 0.81(2) | 2.54(4) | 3.16(8) | 136(5) |
| O(1)–H(10) · N(6) | 0.81(2) | 2.08(3) | 2.82(5) | 13(5) |

Symmetry transformations used to generate equivalent atoms:

1·x+1, y+1, z; ii ·−x, −y, −z; iii ·−x, −y, −z+1; iv ·−x+1, −y, −z; v ·−x+1, −y+1, −z+1.
A paramagnetic Co(II) complex 1 containing an internal standard is added to the inner tube. A solution of the same inert standard, dissolved in the same solvent (DMSO-d6), is placed in the outer tube. The $\Delta f$ values (the frequency separation of the inert reference TMS, in the presence and absence of Co(II) complex) and the molar susceptibility ($\chi_M$) are then calculated. To obtain the corrected $\chi_M$ value, the $\chi_M$ must be corrected by simply summing the diamagnetic contribution (Pascal’s constants) of each ligand atom and group of atoms and then adding it to the susceptibility of the complex [45]. The obtained effective magnetic moments ($\mu_{eff}$) of 4.2 $\mu_B$ for complex 1 is consistent with the expected magnetic moment for octahedral Co(II) complexes (4.3–5.0 BM) for a d$^7$ system with $S = 3/2$ [46].

**TGA studies of [Co(tptz)(CH$_3$OH)Cl$_2$]·CH$_3$OH·0.5H$_2$O (1)**

As is observed in Fig. 6, the TGA/DTA of [Co(tptz)(CH$_3$OH)Cl$_2$]·CH$_3$OH·0.5H$_2$O (1) dried at 100 °C, the total weight loss of the sample is about 75%. The weight loss of the sample from 50 to 100 °C is about 6% which is accompanied with endothermic peaks in its DTA curves. The weight loss in this temperature range is primarily associated with the elimination of physically adsorbed methanol. The sample also shows about 20% weight loss in the 400–520 °C temperature range along with the exothermic peaks in its DTA curves. These peaks are associated with the beginning of weight loss due to the decomposition of some tptz ligand and coordinated methanol [47]. A strong exothermic peak observed at 650 °C in the DTA is associated with the complete removal or decomposition of tptz ligand, shows about 49% weight loss. The final residue is CoCl$_2$ (obs: 24.7% and Calcd: 25%).

**Antibacterial activities**

The in vitro antibacterial activity of tptz and several of its complexes was studied and compared with the activities of two standard antibacterial drugs, viz, Nalidixic acid and Vancomycin. The microorganisms used in this work include *B. subtilis* and *S. aureus* (as Gram-positive bacteria) and *E. coli* and *P. aeruginosa* (as Gram-negative bacteria). Based on the results presented in Table 4 and Fig. 7, it is evident that tptz exhibits moderate activity against *S. aureus*, *B. subtilis* and *P. aeruginosa* with no activity against *E. coli*. Comparison of the antibacterial activities of the ligand and complexes indicates that all complexes exhibit more inhibitory effects than the parent ligand. Such increased activity of the metal chelates could be explained on the basis of chelation theory [48] which emphasizes that chelation tends to make complexes act as more powerful and potent bacteriostatic agents than the free ligands, thus inhibiting the growth of bacteria [49]. In general, permeability across the bacterial cell wall is a prior necessity for the effectiveness of the biocide compounds against different microorganisms. The lipid membrane that surrounds the cell, favors the passage of only lipid soluble materials therefore liposolubility is an important factor that controls antimicrobial activity. On chelation, the overlap of the ligand orbital and partial sharing of the positive charge of the metal ion with the donor

![Fig. 5. Paramagnetic $^1$H NMR of a [Co(tptz)(CH$_3$OH)Cl$_2$]·CH$_3$OH·0.5H$_2$O (1) in DMSO-d$_6$ at room temperature; the assignment of ortho hydrogens for (1) is shown in spectrum.](image)

![Fig. 6. TGA–DTA curves for [Co(tptz)(CH$_3$OH)Cl$_2$]·CH$_3$OH·0.5H$_2$O (1).](image)
groups increases the delocalization of electrons over the whole chelate and enhances the lipophilicity of the complex. The increased lipophilicity in turn enhances the penetration of the complexes into lipid membranes and deactivates various cellular enzymes, which play a vital role on the various metabolic pathways of microorganisms.

Based on the obtained results (Table 4), all metal complexes 1, 2, 3 and 4 were found to inhibit all tested bacteria. It is apparent that the studied complexes are more toxic towards Gram(+) than Gram(-) strains. The reason relies on the difference in the structures of the cell walls and that the Gram(-) cell walls are more complex than those of Gram(+) cells. Lipopolysaccharides form an outer lipid membrane and contribute to the complex antigenic specificity of Gram-negative cells. Particularly significant is that P. aeruginosa was inhibited by complexes while standard drugs were found to exhibit no activity against it.

The antimicrobial activity study revealed that the title compound (1) showed good antibacterial activities against P. aeruginosa strains than the other synthesized compounds with tpptz. It is interesting that P. aeruginosa was inhibited by complexes while, standard drugs were found to have no activity against it. As early proposed cobalt ions have shown to be responsible for the antibacterial activity of cobalt complexes, leading to structural changes in the bacterial cell wall. A thorough investigation regarding the biological effects of the title cobalt complex is essential for medical practice as a metal-based drug. We also tested the antibacterial activity of five metal salts, but they had not any significant inhibitory effects towards bacterial strains.

Geometry optimization of tpptz ligand

Since two different conformers A and B are possible for the tpptz ligand (53) (see supplementary, Scheme S2), the structure of both were fully optimized at the B3LYP method using three basis set including 6-31G, 6-311G and 6-311++G, with no initial symmetry restrictions and assuming C1 point group (Scheme S2). The optimized geometry of the ligand in the gas phase reoptimized by considering the solvent effect (CH3OH) using polarized continuum model (PCM). Tomasi’s polarized continuum model defines the cavity as the union of a series of interlocking atomic spheres. The effect of polarization of the solvent continuum is represented numerically (36). The calculated results indicate that conformer A is about 10 kcal/mol more stable than that of B. The optimized geometry of both conformers of tpptz are shown in Scheme S2, and some structural details in methanol are given in Table S2 (see supplementary).

Table 4
Antibacterial activity data of TPTZ ligand and its complexes.

| Compound | Inhibition zone (mm) |
|----------|----------------------|
|          | E. Coli  | P. aeruginosa | S. aureus | B. subtilis |
| [Co(tpptz)(CH3OH)Cl2] | 12      | 16           | 20    | 21      |
| [MnCl2(tpptz)] (2) | 21      | 12           | 16    | 30      |
| [Ni(tpptz)(CH3OH)Cl2] (3) | 11      | 12           | 12    | 15      |
| [Cu(tpptz)Cl2]nH2O (4) | 17      | 15           | 25    | 26      |
| [Rh(tpptz)Cl3]2H2O (5) | 13      | 14           | 15    | 20      |
| Ligand (6) | n.a. | 11           | 10    | 10      |
| Vancomycin (7) | 13      | n.a.         | 17    | 23      |
| Nalidixic acid (8) | 24      | n.a.         | 12    | 22      |

n.a. = no activity.

Fig. 7. Difference between the antibacterial activities metal complexes (1–5): (1) [Co(tpptz)(CH3OH)Cl2] CH3OH 0.5H2O, (2) [MnCl2(tpptz)], (3) [Ni(tpptz)(CH3OH)Cl2], (4) [Cu(tpptz)Cl2]nH2O, (5) [Rh(tpptz)Cl3] 2H2O with tpptz ligand (6) in comparison with standard antibacterial drugs (7, 8).

Table 5
Selected geometric parameters (Å, °) for the cationic Co(II) complex in 1: experimental (X-ray diffraction – solid phase) and calculated (DFT – different species than in solid phase).

| Geometric parameter | Experimental | Theoretical |
|---------------------|--------------|-------------|
| Co(1)–N(1)          | 2.079(4)     | 2.03        |
| Co(1)–N(4)          | 2.211(4)     | 2.20        |
| Co(1)–N(5)          | 2.218(4)     | 2.21        |
| N(1)–Co(1)–N(4)     | 73.98(14)    | 74.67       |
| N(1)–Co(1)–N(5)     | 74.30(14)    | 74.75       |
| N(4)–Co(1)–N(5)     | 148.14(14)   | 144.69      |
| N(1)–Co(1)–O(1)     | 84.48(12)    | 65.66       |
| N(4)–Co(1)–Cl(1)    | 106.05(10)   | 93.07       |
| N(5)–Co(1)–O(1)     | 87.15(12)    | 83.71       |

Fig. 7. Difference between the antibacterial activities metal complexes (1–5): (1) [Co(tpptz)(CH3OH)Cl2] CH3OH 0.5H2O, (2) [MnCl2(tpptz)], (3) [Ni(tpptz)(CH3OH)Cl2], (4) [Cu(tpptz)Cl2]nH2O, (5) [Rh(tpptz)Cl3] 2H2O with tpptz ligand (6) in comparison with standard antibacterial drugs (7, 8).

Geometry optimization of tpptz ligand

Since two different conformers A and B are possible for the tpptz ligand (53) (see supplementary, Scheme S2), the structure of both were fully optimized at the B3LYP method using three basis set including 6-31G, 6-311G and 6-311++G, with no initial symmetry restrictions and assuming C1 point group (Scheme S2). The optimized geometry of the ligand in the gas phase reoptimized by considering the solvent effect (CH3OH) using polarized continuum model (PCM). Tomasi’s polarized continuum model defines the cavity as the union of a series of interlocking atomic spheres. The effect of polarization of the solvent continuum is represented numerically (36). The calculated results indicate that conformer A is about 10 kcal/mol more stable than that of B. The optimized geometry of both conformers of tpptz are shown in Scheme S2, and some structural details in methanol are given in Table S2 (see supplementary).
Comparison of the theoretical and X-ray data indicates that the standard deviation of 6-31G* and 6-311G** basis sets are similar (Table S2). Therefore, all three methods have good agreement with experimental data. To reduce the computational cost, B3LYP method with 6-31G* basis set was applied to optimize the different complexes.

**Geometry optimization of [Co(tptz)(CH₃OH)Cl₂]·CH₃OH·0.5H₂O (1)**

A model compound of the [Co(tptz)(CH₃OH)Cl₂]·CH₃OH·0.5H₂O complex used in this study was generated from the CIF file. B3LYP method was employed for optimization of cobalt with different high spin (HS) and low spin (LS) states.

The DFT calculations show that the most stable electronic state of [Co(tptz)(CH₃OH)Cl₂]·CH₃OH·0.5H₂O complex is the high spin state (see supplementary Table S3). This result indicates that the ligand field around the Co²⁺ ion is approximately weak. The doublet state is about 8.1 kcal/mol less stable than the quartet spin one.

The optimized structure of [Co(tptz)(CH₃OH)Cl₂]·CH₃OH·0.5H₂O complex with different spin states is shown in Scheme S3 (see supplementary). Results of our calculation at B3LYP/6-31G* are in good agreement with X-ray data. The most relevant geometric parameter in [Co(tptz)(CH₃OH)Cl₂]·CH₃OH·0.5H₂O complex is the cobalt-nitrogen distance. The distances between the Co²⁺ ion and the pyrolic nitrogen are lower than that between the chloride and methanol in the high spin state, (Table 5).

**Calculation of chemical shifts**

The absolute NMR shielding computations were performed using the GIAO method at the DFT optimized structure in the presence of solvent. The ¹H chemical shifts were calculated by using the corresponding absolute shielding calculated for Me₄Si at the same level of theory for both ligand and complex in DMSO as solvent (Table S4, see supplementary). The obtained good agreement between experimental and theoretical chemical shifts shows the reliability of DFT calculations for these series of molecules.

**Molecular orbital analysis point of view**

To find the nature of binding between tptz ligand and Co²⁺, molecular orbital (MO) analysis has been employed. This analysis is useful to present the factors influencing on the stability of these complexes. Taking into account the above discussion, we focused mostly on the [Co(tptz)(CH₃OH)Cl₂] complex in high spin state. The highest occupied molecular orbital (HOMO) and lowest unoccupied molecular orbital (LUMO) of [Co(tptz)(CH₃OH)Cl₂]·CH₃OH·0.5H₂O complex in high spin state are shown in Fig. 8. While the LUMO are nearly based on cobalt 3d orbitals, HOMO includes contributions from both Co²⁺ and interacting ligand heteroatoms. The presented HOMO orbital in Fig. 8 reveals an antibonding interaction between the metal ion and ligand orbitals. These antibonding molecular orbitals are arising from the interaction between the metal orbital and lone pairs of ligand nitrogen atoms.

**Conclusion**

In this study, the complex [Co(tptz)(CH₃OH)Cl₂]·CH₃OH·0.5H₂O (1) was prepared and characterized. The molecular structure of the complex was determined by single-crystal X-ray crystallography. The X-ray structure reveals discrete octahedral coordination geometry for the Co(II) cation. The crystal structure is stabilized by intermolecular H-bonding and π···π stacking interactions. Offset π···π stacking interactions with centroid to centroid separations of 3.672(3) and 3.613(3) Ångstrom are found between the central triazole ring system and the N4 and N6 pyridine rings respectively. The optimized geometric parameters (bond lengths and bond angles) was theoretically determined and compared with the structurally similar compounds. The X-ray structure is in good agreement with its optimized counterpart. The antibacterial activity of metal complexes with the tptz are greater than that of the free ligand. This enhancement in the activity may be due to
increased lipophilicity of the complexes. Whereas \textit{P. aeruginosa} was inhibited by complexes, standard drugs (Vancomycin, Nalidixic acid) were found to be inactive.

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**Appendix A. Supplementary material**

CCDC 934652 contains the supplementary crystallographic data for [Co(tptz)(CH$_3$OH)$_2$Cl]$_2$CH$_3$OH•0.5H$_2$O (1). 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. Supplementary data associated with this article can be found, in the online version, at http://dx.doi.org/10.1016/j.molstruc.2015.03.037.

**References**

[1] M. Chauhan, K. Banerjee, F. Arjmand, Inorg. Chem. 46 (2007) 3072–3082.
[2] M.N. Patel, P.A. Dosi, B.S. Bhattacharya, Spectrochim. Acta A 68 (2012) 508–514.
[3] Z.A. Siddiqui, M. Khalid, S. Kumar, S. Shadid, S. Noor, Eur. J. Med. Chem. 45 (2010) 264–269.
[4] A.T. Kabahani, H.H. Hammud, A.M. Ghannoun, Chem. Pharm. Bull. 55 (2007) 446–450.
[5] M. Scarpellini, A. Neves, R. Horner, A.J. Bortotulluzi, C. Szpoganics, C. Zucco, R.A. Nome Silva, V. Drago, A.S. Mazzarich, W.A. Ortiz, W.A. Passos, M.C. Oliveira, H. Rodriguez-Monge, R. Taylor, J. von der, S.P.A. Wood, J. Appl. Crystallogr. 41 (2008) 466–470.
[6] H. Chao, L.-N. Ji, Y. Cao cobalt complexes as potential pharmaceutical agents, in: M. Gielen, E.R.T. Kessissoglou, G. Psomas, Dalton Trans. 39 (2010) 4517–4528.
[7] P. Kumar, A.K. Singh, J.K. Saxena, D.S. Pandey, J. Organomet. Chem. 694 (2009) 3570–3579.
[8] V.P. Singh, A. Katiyar, S. Singh, Biometals 21 (2008) 491–501.
[9] T. Takeuchi, A. Bottcher, C.M. Quezada, T.J. Meade, H.B. Gray, Bioorg. Med. Chem. 23 (2005) 3570–3579.
[10] M. Maghami, F. Farzaneh, J. Simpson, A. Moazeni, Polyhedron 73 (2014) 22–29.
[11] A. Bury, A.E. Underhill, D.R. Kemp, N.J. Oshea, J.P. Smith, P.S. Gomm, Inorg. Chim. Acta 416 (2014) 109–118.
[12] R. Maccari, R. Ottana, B. Bottari, E. Rotondo, M.G. Vigorita, Bioorg. Med. Chem. Lett. 14 (2004) 1901–1904.
[13] P. Paul, B. Tyagi, A.K. Bilakhiya, M.M. Bhadbhade, E. Suresh, J. Chem. Crystallogr. 42 (2012) 656–667.
[14] K. Abdi, H. Hadadzadeh, M. Weil, H. Amiri, Inorg. Chim. Acta 416 (2014) 109–118.
[15] K. Ha, Acta Cryst. E66 (2010) m262.
[16] P. Paul, B. Tyagi, A.K. Bilakhiya, M.M. Bhadbhade, E. Suresh, J. Ramachandraiah, Inorg. Chem. 37 (1998) 5733–5742.
[17] J.H. McKerrow, E. Sun, P.J. Rosenthal, J. Bouvier, Annu. Rev. Microbiol. 47 (1993) 821–853.
[18] K. Nakamoto, Infrared and Raman Spectra of Inorganic and Coordination Compounds: Part II. Application in Coordination, Organometallic and Bioinorganic Chemistry, sixth ed., Wiley-Interscience, New York, 2009.
[19] H. Hadadzadeh, G. Mansouri, A. Rezvani, H.R. Khavasi, B.W. Skelton, M. Makha, F.R. Chazari, Polyhedron 30 (2011) 2355–2364.
[20] H. Zhao, M. Shatruk, A.V. Prosvirin, K.R. Dunbar, Chem. Eur. J. 13 (2007) 6573–6580.