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Synthesis, Characterization and *in vitro* Antimicrobial Activity of Organotellurium Decorated 10-Membered Tetraazamacrocyclic Complexes of Cobalt(II)

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Three novel organotellurium decorated 10-membered tetraazamacrocyclic complexes of Co(II) were synthesized using template condensation of 1,1-diiodo-1-telluracyclopentane, 1,1-diodo-2-methyl-1-telluracyclopentane and 1,1-diiodo-1,1-diethyltellurium(IV) with 1,2-diaminopropane and cobalt dichloride hexahydrate in dry methanol in 2:2:1 molar ratio. The characterization of the synthesized cobalt(II) complexes was carried out using elemental analysis, spectroscopic studies (IR, proton NMR & proton decoupled carbon NMR) and molar conductance measurements. The results of these studies suggested that the complexes may be formulated as [$CoLCl_2$] where L = $\{C_4H_8Te(NHCH_2CH(CH_3)NH)\}_2$, $\{C_4H_7(CH_3)Te(NHCH_2CH(CH_3)NH)\}_2$ and $\{(C_2H_5)_2Te(NHCH_2CH(CH_3)NH)\}_2$. The synthesized complexes were also screened for antimicrobial activity using broth microdilution and agar disc diffusion methods.

Keywords: Organotellurium, Macrocyclic complexes, Spectroscopic studies, Antimicrobial activity.

INTRODUCTION

The design and the synthesis of metal containing macrocyclic compounds is a field of great interest [1]. Synthetic macrocyclic complexes can mimic some naturally occurring macrocycles due to their resemblance with natural macrocycles, such as metalloproteins and metalloenzymes [2,3]. Macrocyclic complexes have applications in the field of biometallic activation and catalysis [4], pharmaceuticals [5] and pigments and dyes [6]. Macrocyclic compounds possess very good selective complexing properties [7,8] and their metal complexes are used as catalysts [9-11]. Since, the macrocyclic complexes having soft and hard donors with a metal center play a significant role in MOCVD processes [12-14] and transition metal-catalyzed asymmetric synthesis [15,16].

The metal complexes of macrocyclic ligands containing tellurium as soft donor and N and O as hard donors have gained great attention and attempts have been made to explore their chemistry [17-19]. Synthesis of transition metal complexes of tellurium containing tetraazamacrocycles by template method has been reported [20,21]. Kumari *et al.* [22] reported the synthesis of divalent transition metal complexes of 10-membered tellurium containing dithiadiazamacrocycles. Synthesis of biologically active Ni(II), Pd(II) and Pt(II) complexes of tellurium containing tetraazamacrocycles are also reported in the literature [23]. In light of the above facts, in the present communication, the synthesis, spectral study and antimicrobial activity of three novel organotellurium decorated 10-membered tetraazamacrocyclic complexes of Co(II) are reported.

EXPERIMENTAL

The chemical reagents and the solvents employed in the synthesis were of analytical grade and procured commercially from Sigma-Aldrich and Merck, USA. The organic solvents were properly dried and distilled before use. IR spectra were recorded using Agilent Cary 630 FTIR spectrometer in the frequency range 4000-400 cm^{-1}. The ^{1}H and ^{13}C{^{1}H} NMR spectra were recorded at 300 MHz using Bruker Avance 400/Avill HD-300 (FT NMR) spectrometer in DMSO containing tetramethyl silane as an internal standard. Elemental analysis for H, C and N were performed on Euro Vector Elemental Analyzer. The tellurium and chlorine contents were determined in the laboratory volumetrically and cobalt [24] content was estimated gravimetrically. Measurement of molar conductance was carried out on Systronic type 305 Conductivity Bridge in DMSO solvent using a dip-type cell with a smooth platinum electrode. Melting points were recorded in open capillary and are uncorrected. The antimicrobial activity was studied using the broth microdilution and disc diffusion methods. Nutrient broth (NB), yeast extract peptone dextrose (YEPD), Mueller Hinton agar (MHA) and broth were purchased from HiMedia Lab. Pvt. Ltd., India. Bacterial and fungal strains were provided by the Department of Microbiology, Central University of Punjab, Bathinda, India.

Synthesis of organotellurium(IV) diiodides: 1,1-Diiodo-1-telluracyclopentane [$C_4H_8TeI_2$] [25], 1,1-diodo-2-methyl-1-telluracyclopentane [$C_4H_7(CH_3)TeI_2$] [26] and 1,1-diiodo-1,1-diethyltellurium (IV) [$(C_2H_5)_2TeI_2$] [27] were synthesized by the reaction of tellurium metal with 1,4-diiodobutane, sodium iodide and 1,4-dibromopentane and ethyl iodide, respectively, by the methods available in the literature.

Synthesis of cobalt(II) complexes of organotellurium decorated 10-membered tetraazamacrocycles

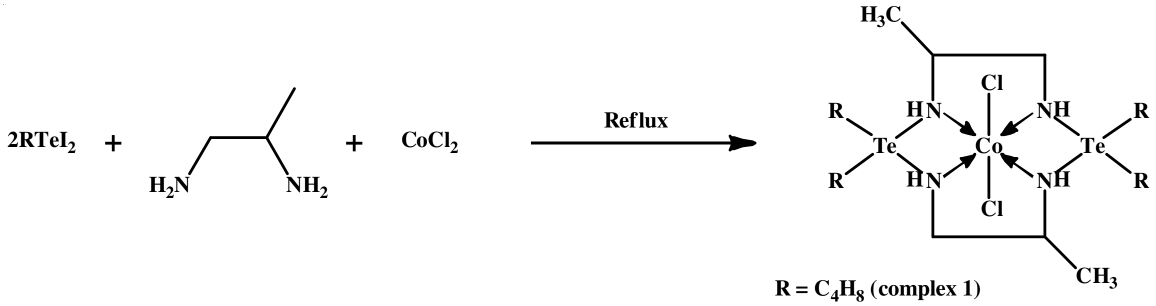
Synthesis of [$Co\{(C_4H_8Te)(NHCH_2CH(CH_3)NH)\}_2Cl_2$] (1): A saturated solution of 1,1-diiodo-1-telluracyclopentane (1.0 g, 2.28 mmol) in methanol was added dropwise with constant stirring to a methanolic solution (5 mL) of 1,2-diaminopropane (2.28 mmol). The colour of the reaction mixture was changed from orange to light yellow and then refluxed for 4 h thereafter a solution of $CoCl_2·6H_2O$ (0.276 g, 1.16 mmol) in methanol was added. A clear change in colour was obtained, however, the refluxing was further continued for 8 h. The reaction mixture was then cooled to room temperature and filtered. The filtrate was concentrated at reduced pressure and allowed for slow evaporation for crystallization. After 3 days, the obtained crystalline solid was washed with petroleum ether and finally dried (**Scheme-I**). Light orange crystal, yield: 33.88%, m.p.: 160 ºC, Elemental analysis of $C_{14}H_{32}Cl_2CoN_4Te_2$, calcd. (found) %: C, 26.21 (26.39); H, 5.03 (5.17); N, 8.73 (8.60); Cl, 11.05 (11.24); Co, 9.19 (8.95); Te, 39.78 (39.60). Molar conductance: 19.40 Ω^{-1} cm^2 mol^{-1}. IR (KBr, ν_{max}, cm^{-1}): 2910 (C-H); 1630 (N-H), 1160 (C-N), 815 (N-H), 525 (Te-C), 465 (Co-N), 414 (Te-N), 315 (Co-Cl). ^{1}H NMR (δ ppm): 2.47 (s, 4H, NH), 2.65-3.27 (m, 6H, CH,CH_2, propylenic chain), 1.35 (d, 6H, aliphatic CH_3), 4.03 (m, 8H, $TeCH_2$), 3.32 (m, 8H, $TeCCH_2$); $^{13}C\{^1H\}$ NMR (δ ppm): 45.42 ($TeCH_2$), 32.69 ($TeCCH_2$), 49.35 (NCH_2), 48.80 (NCH), 17.23 ($NCCH_3$).

Synthesis of [$Co\{(C_4H_7(CH_3)Te)(NHCH_2CH(CH_3)NH)\}_2Cl_2$] (2): Complex **2** was synthesized by the reaction of 1,1-diiodo-2-methyl-1-telluracyclopentane (1.0 g, 2.21 mmol) with 1,2-diaminopropane (2.21 mmol) and $CoCl_2· 6H_2O$ (0.263 g, 1.106 mmol) by following the above mentioned procedure in the similar manner (**Scheme-I**). Light orange crystal, yield: 69.73%, m.p.: 218 ºC, Elemental analysis of $C_{16}H_{36}Cl_2CoN_4Te_2$, calcd. (found) %: C, 28.70 (28.63); H, 5.42 (5.31); N, 8.37 (8.50); Cl, 10.59 (10.37); Co, 8.80 (8.96); Te, 38.12 (38.33). Molar conductance: 16.70 Ω^{-1} cm^2 mol^{-1}. IR (KBr, ν_{max}, cm^{-1}): 2919 (C-H); 1639 (N-H), 1153 (C-N), 826 (N-H), 546 (Te-C), 472 (Co-N), 421 (Te-N), 325 (Co-Cl). ^{1}H NMR (δ ppm): 2.39 (s, 4H, NH), 2.50-3.12(m, 6H, CH,CH_2, propylenic chain), 1.37 (d, 6H, aliphatic CH_3), 5.04-4.57 (m, 6H, TeCH, $TeCH_2$), 4.48-4.27 (m, 8H, $TeCCH_2$), 1.27 (d, 6H, $TeCCH_3$); $^{13}C\{^1H\}$ NMR (δ ppm): 53.16 ($TeCH_2$), 51.81 (TeCH), 48.79, 49.56 ($TeCCH_2$), 16.97 ($TeCCH_3$), 17.19 ($NCCH_3$), 49.19 (NCH_2), 48.81 (NCH).

Synthesis of [$Co\{((C_2H_5)_2)Te)(NHCH_2CH(CH_3)NH)\}_2Cl_2$] (3): Similarly, complex **3** was also synthesized by reacting 1,1-diiodo-1,1-diethyltellurium (IV) (1.0 g, 2.27 mmol) with 1,2-diaminopropane (2.27 mmol) and $CoCl_2· 6H_2O$ (0.27 g, 1.14 mmol) (**Scheme-I**). Light orange crystal, yield: 61.96%, m.p.: 210 ºC, Elemental analysis of $C_{14}H_{36}Cl_2CoN_4Te_2$, calcd. (found) %: C, 26.05 (26.14); H, 5.62 (5.53); N, 8.68 (8.54); Cl, 10.98 (11.13); Co, 9.13 (9.25); Te, 39.54 (39.41). Molar conductance: 17.10 Ω^{-1} cm^2 mol^{-1}. IR (KBr, ν_{max}, cm^{-1}): 2913 (C-H); 1632 (N-H), 1158 (C-N), 816 (N-H), 526 (Te-C), 466 (Co-N), 417 (Te-N), 316 (Co-Cl). ^{1}H NMR (δ ppm): 2.34 (s, 4H, NH), 2.57-3.19 (m, 6H, CH,CH_2, propylenic chain), 1.34 (d, 6H, aliphatic CH_3), 2.96 (q, 8H, $TeCH_2$), 0.85 (t, 12H, $TeCCH_3$); $^{13}C\{^1H\}$ NMR (δ ppm): 29.11($TeCH_2$), 8.14 ($TeCCH_3$), 17.27 ($NCCH_3$), 49.17 (NCH_2), 48.75 (NCH).

Antimicrobial study

Broth microdilution method: The broth microdilution method [28] was used to examine the antimicrobial efficacy of synthesized organotellurium macrocyclic complexes of Co(II) (**1-3**). The bacterial cells were inoculated in nutrient broth (NB) medium at 37 ºC and the fungal cells in yeast extract peptone dextrose (YEPD) were grown at 30 ºC with shaking overnight. The overnight grown culture was sub-cultured in sterile NB/



R = C_4H_8 (complex 1)
$C_4H_7(CH_3)$ (complex 2)
$(C_2H_5)_2$ (complex 3)

Scheme-I: Synthesis of Co(II) complexes of tellurium containing tetraaza macrocycles

YEPD broth till OD_{600} nm reached 0.4 (TECAN, Spark). The cells were diluted in 1:1000 ratios in MH broth to reach the 1×10^6 CFU/mL bacterial culture density. A volume of 100 µL of this bacterial solution was poured into 96-wells of sterile culture plate and mixed with an equal volume of two-fold serially diluted with a starting concentration of 200 µg/mL of each **1**, **2** and **3**. The bacterial growth was visually observed after 16 h of incubation.

Disc diffusion method: The disc diffusion method also was used to examine the antimicrobial activity of complexes **1**, **2** and **3**. In brief, bacterial cells (*E. coli*, *S. aureus*, *K. peumoniae* and *P. aeruginosa*) were inoculated in a nutrient broth (NB) medium thereafter incubated overnight at 37 ºC at 200 rpm (Orbital shaking incubator, Creative lab world). The overnight grew culture was sub-cultured (1%) in NB broth and the fungal cells (*C. albicans*) in YEPD were grown at 30 ºC with shaking overnight and incubated till the optical density (OD) reached 0.4 at 600 nm [29,30]. Meanwhile, Muller-Hilton agar (MHA) media was poured into a sterile petridish (90 mm × 15 mm, Genaxy, India) and kept for solidification. A 150 µL of bacterial and fungal culture (OD_{600} nm 0.4) were spread over the MHA plate. Then, the sterile blank discs were placed over MHA plates. Further, the blank sterile discs were wetted with 40 µL of concentration 200 µg/mL of each of the three complexes (**1**, **2** and **3**) and a sterile blank disc was wetted with distilled water for negative control and ciprofloxacin of 5 µg for bacterial cells and itraconazole of 10 mcg for fungal cells was used as a positive control. These plates were incubated at 37 ºC/30 °C overnight and after 14 h of incubation, the zone of inhibition was analyzed.

RESULTS AND DISCUSSION

Three organotellurium(IV) diiodides, *viz.* 1,1-diiodo-1-telluracyclopentane [$C_4H_8TeI_2$], 1,1-diodo-2-methyl-1-telluracyclopentane [$C_4H_7(CH_3)TeI_2$] and 1,1-diiodo-1,1-diethyltellurium(IV) [$(C_2H_5)_2TeI_2$] when refluxed with 1,2-diaminopropane and cobalt chloride hexahydrate in 2:2:1 molar ratio produced 10-membered tetraazamacrocyclic complexes **1**, **2** and **3**, respectively. The proposed structures for these complexes are given in Fig. 1. All the synthesized cobalt (II) complexes were soluble in most of the organic solvents like benzene, methanol, DMF and DMSO and insoluble soluble in waterThe low molar conductance (Λ_m) values for complexes **1-3** in DMSO solvent predicts that these complexes are non-electrolytes in nature. The analytical data and the low Λ_m values suggest the complexes have a general formula [$CoLCl_2$] where L is organotellurium decorated tetraazamacrocycle.

Spectral studies: In the synthesized organotellurium macrocyclic cobalt(II) complexes (**1-3**), the absence of the IR bands corresponding to the free NH_2 group and the presence of medium intensity bands at 1639-1630 cm^{-1} and 826-815 cm^{-1} assigned to NH deformation and NH out of plane bending vibrations, respectively, which strongly approves the formation of macrocyclic skeleton [31]. The formation of a Te-containing macrocycle is further supported by the appearance of a new weak band at 421-414 cm^{-1} attributed to Te-N vibrations [21,30]. The presence of medium intensity bands at 1160-1153 and 472-465 cm^{-1} corresponds to C-N and Co-N vibrations, respectively [21,31,32]. Further medium intensity bands at 2919-2910 cm^{-1} and weak bands at 325-315 cm^{-1} appeared due to C-H and Co-Cl vibrations, respectively [33-35].

The proton NMR spectra of 1,2-diaminopropane exhibits peaks at 0.45 ppm, 1.25 ppm, 1.67 ppm and 1.83 ppm which correspond to amino, methyl, methylene and methine groups, respectively [30,36]. In ^1H NMR spectra of complexes **1-3** peak corresponding to the primary amino group is absent and a broad singlet appeared at δ 2.34 -2.47 ppm due to the coordinated 2º amino group [30]. The CH_2 protons, present in the propylenic chain, appeared at δ 1.34-1.37 ppm as doublets, whereas the methylene and methine protons appeared at δ 2.65-3.12 ppm as multiplet. This shows that these protons are deshielded due to coordination of nitrogen with metal. In NMR spectra of complex **1**, $TeCH_2$ and $TeCCH_2$ protons appeared at δ 4.03 ppm and 3.32 ppm as multiplets. In complex **2**, TeCH and $TeCH_2$ protons appeared at δ 5.04-4.57 ppm as multiplet, similarly $TeCCH_2$ protons appeared at δ 4.48-4.27 ppm as multiplet, whereas $TeCCH_3$ protons appeared at δ 1.27 ppm as doublet. The proton NMR spectra of complex **3** contained a quartet at δ 2.96 ppm and a doublet at δ 0.85 ppm due to $TeCH_2$ and $TeCCH_3$ protons, respectively. In carbon NMR spectra of the complexes **1-3**, methyl, methylene and methine carbons appeared in the range δ 17.19-17.27 ppm, 49.17-49.35 ppm and 48.75-48.81 ppm, respectively as singlets. In complexes **1**, **2** and **3**, $TeCH_2$ carbon appeared at δ 45.42 ppm, 53.16 ppm & 51.81 ppm and 29.11 ppm, respectively. In **1** and **2**, $TeCCH_2$ appeared at δ 32.69 ppm and δ 48.79 & 49.56 ppm, respectively. In complexes **2** and **3**, $TeCCH_3$ carbon singlet appeared at δ 16.97 ppm and 8.14 ppm, respectively. Thus, the NMR peaks are in good agreement with the proposed molecular structures for the complexes and support the formation of a macrocyclic skeleton containing organotellurium moiety.

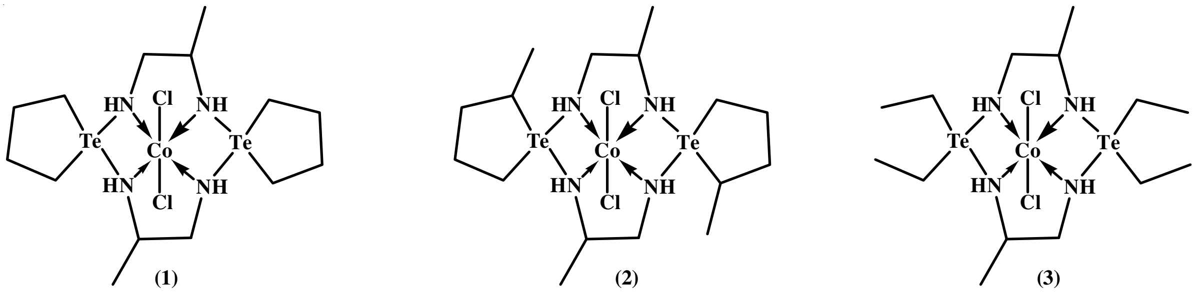


Fig. 1. Molecular structures of Co(II) complexes of tellurium containing tetraaza macrocycles

Antimicrobial activity: The antimicrobial inhibitory efficacy of complexes **1**, **2** and **3** was examined using the broth microdilution method. The antimicrobial activity data (Table-1) suggested that complexes **1**, **2** and **3** inhibited bacterial strains *E. coli, S. aureus, K. pneumoniae* and *P. aeruginosa* and fungal strain *C. albicans* with specific MIC values among the concentration gradients. MIC of compound **1** against *E. coli* is 50 µg/mL whereas against *S. aureus, K. pneumoniae*, *P. aeruginosa* and *C. albicans* it is 100 µg/mL. The MIC value of compound **2** against all the tested bacterial and fungal strains is 200 µg/mL. *S. aureus* is 200 µg/mL, *K. pneumoniae* is 200 µg/mL, *P. aeruginosa* is 200 µg/mL and *C. albicans* is 200 µg/mL. The MIC of complex **3** against *E. coli, S. aureus* and *P. aeruginosa* is 200 µg/mL whereas against *K. pneumoniae* and *C. albicans* it is 100 µg/mL.

The efficacy of complexes **1**-**3** was further studied using the disc diffusion method. Diffusion of compounds into MHA media prompts hindrance of bacterial and fungal growth and forming of a clear zone of inhibition (ZOI) around the discs. Initially at lower concentration-based inhibition of bacterial and fungal growth was observed against *E. coli*, *S. aureus*, *K. pneumoniae*, *P. aeruginosa* and *C. albicans*, but at 200 µg/mL. Distilled water was the negative control, ciprofloxacin (5 mcg) for bacterial cells and itraconazole (10 mcg) for fungal was used as a positive control. The ZOI value for complexes **1**, **2** and **3** against the studied strains are shown in Table-2, which clearly showed the antibacterial and antifungal effects and significant inhibition.

Conclusion

The synthesis, spectroscopic and antimicrobial study of novel organotellurium decorated tetraazamacrocyclic complexes of Co(II) have been described. The complexes have been synthesized by template condensation of organotellurium diiodides with 1,2-diaminopropane and cobalt chloride hexahydrate in 2:2:1 molar ratio. The antimicrobial activity among all three complexes (**1**, **2** and **3**) only complex **1** showed good antimicrobial activity against all the tested bacterial/fungal strains.

CONFLICT OF INTEREST

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

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TABLE-1
MIC VALUES OF COMPLEX **1**, **2** AND **3** AGAINST BACTERIAL AND FUNGAL PATHOGEN STRAINS

Strain	Complex **1**						Complex **2**						Complex **3**					
	200	100	50	25	12.5	6.25	200	100	50	25	12.5	6.25	200	100	50	25	12.5	6.25
	µg/mL						µg/mL						µg/mL					
E. coli	–	–	–	+	+	+	–	+	+	+	+	+	–	+	+	+	+	+
S. aureus	–	–	+	+	+	+	–	+	+	+	+	+	–	+	+	+	+	+
K. pneumoniae	–	–	+	+	+	+	–	+	+	+	+	+	–	-	+	+	+	+
P. aeruginosa	–	–	+	+	+	+	–	+	+	+	+	+	–	+	+	+	+	+
C. albicans	–	–	+	+	+	+	–	+	+	+	+	+	–	-	+	+	+	+

"–" sign means the absence of bacterial growth at that concentration; "+" sign means the presence of bacterial growth at that concentration.

TABLE-2
ZONE OF INHIBITIONS (mm) VALUES OF COMPLEX **1**, **2** AND **3** AGAINST BACTERIAL AND FUNGAL PATHOGEN STRAINS

Compounds (200 µg/mL)	Bacterial strains				Fungal strain
	E. coli	*S. aureus*	*K. pneumoniae*	*P. aeruginosa*	*C. albicans*
1	14	15	20	10	17
2	10	09	00	00	12
3	10	10	10	00	10
Ciprofloxacin (5 mcg)	19	23	25	35	–
Itraconazole (10 mcg)	–	–	–	–	19

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