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Research Article

Spectroscopic Characterization, *In Vitro* Cytotoxicity, and Antioxidant Activity of Mixed Ligand Palladium(II) Chloride Complexes Bearing Nucleobases

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Mixed-ligand palladium(II) chloride complexes bearing the nucleobases, adenine (Ad), cytosine (Cyt), and guanine (Gua), have been synthesized and characterized by UV-vis spectrophotometric methods, magnetic susceptibility, molar conductivity, elemental analysis, FTIR, and ^{1}H-NMR. The complexes were found to have the composition cis-[$PdCl_2$(Gua)(Cyt)], cis-[$PdCl_2$(Ad)(Cyt)], and cis-[$PdCl_2$(Ad)(Gua)]. A four-coordinated square-planar geometry is proposed for these Pd(II) complexes based on magnetic evidence and electronic spectra. The complexes as well as the free nucleobase ligands were tested for their *in vitro* cytotoxicity on human promyelocytic leukemia (HL60) and human histiocytic leukemia (U937) cell lines. cis-[$PdCl_2$(Ad)(Gua)] showed IC_{50} values of 11.29 ± 2.91 and $8.31 \pm 1.44\ \mu$M against HL60 and U937, respectively, which was higher than that of the positive control (curcumin) against U937. The complexes also showed significant antioxidant activity when tested against 2,2-diphenyl-1-picrylhydrazylradical (DPPH).

1. Introduction

The versatility of nucleobases has made them suitable ligands for the synthesis of numerous transition metal complexes [1–5]. In particular, palladium complexes of nucleobases and their derivatives have been of current interest since these complexes form faster than their platinum analogues and produce analogous products in solution [6, 7]. The mode of bonding and structure of these and related ligands have been exhaustively studied, both spectroscopically and crystallographically [8–11] and have been shown to be monodentate via the N3 of the pyrimidine and N7 of the purine ligand (as shown in Figure 2). In view of the strong electron withdrawing ability of these metal ions, there is a drift of electron density onto the metal centre and in effect ligand hydrogens are easily lost in the presence of a free radical. This makes these metal complexes act as better antioxidants as compared to the free ligand [12, 13].

Mixed-ligand complexes play a key role in biological chemistry [14] because mixed chelation occurs commonly in biological fluids as millions of potential ligands are likely to compete for metal ions *in vivo* [15]. Because of this profound role, mixed-ligand complexes have been extensively studied for their thermodynamic [16–18] as well as kinetic stability [19–23]. These complexes have received tremendous attention in the search for novel drugs against drug resistant diseases [24–27] with cisplatin being a classic example [28].

Metal based antioxidants have received recent attention for their capacity to protect organisms and cells from damage induced by oxidative stress [12]. An effective antioxidant, however, should be able to terminate the attack of reactive species like free radicals and prevent them from attacking body cells. The antioxidant activity of a synthetic compound can be measured using the scavenging ability of that compound to trap free radicals [13]. Although some palladium(II) complexes of pyrimidine and pyridine derivatives have been synthesized and characterized [1, 29], not much has been reported about the antioxidant activity as well as the *in vitro* cytotoxicity of these complexes. In this study we present the synthesis, spectroscopic characterization, and

antioxidant as well as *in vitro* cytotoxicity of palladium(II) complexes with purine and pyrimidine nucleobases as ligands on human promyelocytic leukemia (HL60) and human histiocytic leukemia (U937) cell lines.

2. Materials and Methods

2.1. Materials. All the ligands were purchased from Molekula (UK) and the palladium(II) chloride was purchased from Merck KGaA (Germany). These were used without further purification. The DPPH was purchased from Sigma-Aldrich (St. Louis, MO, USA). All other chemicals were of analytical grade and obtained from standard suppliers. Palladium content was analysed spectroscopically using dimethylglyoxime and a modification of the method described by Khader and Prasad [30]. Free radical scavenging ability of the complexes was used as a measure of the antioxidant activity of the complexes. Thiazoyl blue tetrazolium bromide (MTT) assay was used to determine the cytotoxicity of the complexes against U937 and HL60 cell lines.

2.2. Instrumentation and Methods. Chloride content was determined by the Mohr method. ^{1}H-NMR was recorded in DMSO-d_6 on a Gemini 2000 instrument (400 MHz) at room temperature and the ^{1}H chemical shifts referenced to the residual signals of the protons of the NMR solvent quoted in ppm. The IR spectra (KBr disks) were recorded on an Interspec 200-X spectrophotometer. Molar conductivity was recorded on a Wagtech 4510 conductivity meter. Electronic absorbance of the complexes was recorded in DMSO on a T70 UV/VIS spectrometer. Melting points of the complexes were determined with a Thermo Scientific Electrothermal Digital Melting Point Apparatus IA9100. Magnetic susceptibility was determined using a modification of the Gouy method [31, 32].

2.3. Synthesis of the Palladium(II) Complexes. The complexes were prepared in a general way as follows: to a 10% HCl ethanolic solution (10 mL) of $PdCl_2$ (0.354 g, 2 mmol) was added 2 mmol of one nucleobase ligand in 20 mL of boiling ethanol. After stirring the mixture at 100°C for 30 minutes, 2 mmol of the other ligand dissolved in 20 mL of boiling ethanol was then added. The resulting solution was refluxed at 100°C with stirring for further 45 minutes. The yellow to orange precipitates were suction-filtered and washed with hot ethanol and dried over anhydrous $CaCl_2$.

2.4. Procedure for MTT In Vitro Cytotoxicity. The *in vitro* cytotoxicity of the compounds was performed on HL60 and U937 cells as described [33]. The cells were maintained in RPMI 1640 medium (containing 10% FBS and 0.01% kanamycin). Cells in exponential growth were seeded into 96-well plates at a concentration of 1×10^4 cells/100 μL/well. The cells were then treated with various concentrations of the test compounds at a concentration range of 0–200 μM for HL60 and 0–100 μM for U937 using DMSO as the solvent. Negative control (untreated) and positive control (curcumin) experiments were included.

After 72 hours of incubation at 37°C under 5% CO_2 in humidified atmosphere, 20 μL of MTT (3-(4,5-dmethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide) was added to each well and the plates were kept in the dark for further 4 hours. Subsequently, 150 μL of acidified isopropanol containing Triton-X 100 was added to stop the reaction and also solubilize the formazan crystals formed. Absorbance readings were taken at 570 nm on a Tecan-PC infinite M200 Pro Plate reader after overnight incubation of the plates. Triplicate experiments were performed. Dose response curves were plotted as percentages of cell viability against concentration. Drug sensitivity was expressed in terms of the concentration of drug required for a 50% reduction of cell viability (IC_{50}). The IC_{50} values were determined by nonlinear regression analysis.

2.5. Procedure for Antioxidant Activity. The antioxidant activity of the complexes was determined as described [12, 34] with slight modifications as follows: on a 96 well plate, the compounds were serially diluted in DMSO to obtain a concentration range of 1.25–20 mM. The reaction mixture consisted of 100 μL of 0.5 mM 2,2-diphenyl-1-picrylhydrazylradical (DPPH), and 100 μL of each concentration of the test compounds using DMSO as the solvent. For positive control, 2,6-di-tert-butyl-4-methylphenol (BHT) was used at a concentration range of 0.0625–2 mM in methanol. The solvents, DMSO and methanol, were used as blanks. Duplicate experiments were performed. The plates were covered with aluminum foil and kept in the dark for 20 minutes after which the absorbance was read on a Tecan-PC infinite M200 Pro Plate reader at the absorbance wavelength of 517 nm. The percent antioxidant activity was calculated as follows:

$$\left[\text{Percent antioxidant} = \frac{A_0 - A_1}{A_0}\right], \quad (1)$$

where A_0 is the absorbance of the blank and A_1 is the absorbance in the presence of the sample or positive control.

3. Results and Discussion

3.1. cis-[$PdCl_2$(Gua)(Cyt)]. Yield = 0.6905 g (78.5%). M.p (dec) = 250°C. Colour = yellowish brown. Anal. Calcd. for $C_9H_{10}N_8O_2PdCl_2$ (%): Pd, 24.21; Cl, 16.16. found: Pd, 23.91; Cl, 15.81. UV-Vis λ (nm) (ε): 286(19007 M^{-1} cm^{-1}), 331(3819 M^{-1} cm^{-1}), 403(871 M^{-1} cm^{-1}), 531(624 M^{-1} cm^{-1}). Λ_m (R^2): 13.58 Ω^{-1} cm^{-2} mol^{-1} (0.987). μ_{eff} = 0.

3.2. cis-[$PdCl_2$(Ad)(Cyt)]. Yield = 0.6905 g (81.5%). M.p (dec) = 250°C. Colour = yellowish brown. Anal. Calcd. for $C_9H_{10}N_8OPdCl_2$(%): Pd, 26.13; Cl, 16.77. found: Pd, 26.10; Cl, 16.12. UV-Vis λ (nm) (ε): 280(5149 M^{-1} cm^{-1}), 331(2326 M^{-1} cm^{-1}), 406(1111 M^{-1} cm^{-1}), 531(503 M^{-1} cm^{-1}). Λ_m (R^2): 18.48 Ω^{-1} cm^{-2} mol^{-1} (0.983). μ_{eff} = 0.

3.3. cis-[$PdCl_2$(Gua)(Ad)]. Yield = 0.6812 g (73.5%). M.p (dec) = 250°C. Colour = yellowish brown. Anal. Calcd. for $C_{10}H_{10}N_{10}OPdCl_2$(%): Pd, 22.96; Cl, 15.32. found: Pd,

Table 1: Major IR/cm^{-1} absorption bands of the palladium(II) complexes and the nucleobase ligands.

Complex	ν_{as} (NH_2)	ν_s (NH_2)	ν_s (C=O)	ν_s (C=N)	δ_s (NH_2)	H-bonding
cis-[$PdCl_2$(cyt)(gua)]	3424 w	3286 s	1705 s	1634 s	1605 m	3625–2662 br
cis-[$PdCl_2$(cyt)(ad)]	3416 w	3271 w	1672 s	1638 w	1590 m	3663–2800 br
cis-[$PdCl_2$(cyt)(gua)]	3323 w	3170 w	1698 s	1638 s	1567 m	3618–2587 br
Cytosine	3372 w	3166 w	1668 s	1620 s	1534 m	3500–2500 br
Adenine	3364 w	3230 w	—	1672 s	1601 m	3394–2500 br
Guanine	3308 w	3114 w	1705 s	1631 s	1563 m	3390–2500 br

ν_{as}: antisymmetric stretching, ν_s: symmetric stretching, δ_s: symmetric bending, s: strong, w: weak, br: broad, and m: medium.

Table 2: Far IR/cm^{-1} bands for the palladium(II) complexes.

Complex	ν_s (Pd–Cl)	ν_s (Pd–N)
cis-[$PdCl_2$(cyt)(gua)]	389 w, 312 w	282 w, 238 w
cis-[$PdCl_2$(cyt)(ad)]	386 w, 311 w	281 w, 227 w
cis-[$PdCl_2$(cyt)(gua)]	364 w, 339 w	283 w, 248 w

22.85; Cl, 15.33. UV-Vis λ (nm) (ε): 290(14001 M^{-1} cm^{-1}), 333(3071 M^{-1} cm^{-1}), 407(784 M^{-1} cm^{-1}), 531(761 M^{-1} cm^{-1}). Λ_m (R^2): 15.32 Ω^{-1} cm^{-2} mol^{-1} (0.925). $\mu_{eff} = 0$.

3.4. IR Spectra. Important vibrational bands were selected by comparing the FTIR spectra of the complexes with those of the respective free nucleobase ligands. The symmetric and antisymmetric N–H stretching frequencies of the complexes were observed in the range of 3424–3170 cm^{-1}. These peaks were compared with those of the free ligands which appear between 3372 and 3114 cm^{-1}. The observed shifts can be attributed to hydrogen bonding and other noncovalent interactions in the solid state of these metal complexes [35, 36]. These observations are supported by the broadening of the spectra observed at 3663–2500 cm^{-1} [36]. Also observed around 1605–1567 cm^{-1} are important symmetric NH_2 bending vibrations of the ligands which showed slight shifts as compared to those of the free ligands. Furthermore, peaks corresponding to the ring stretching frequencies ν_s C=N between 1620–1567 cm^{-1} in the free ligands were found to have shifted to 1634–1638 cm^{-1} in the metal complexes, suggesting the involvement of the ring C=N group in coordinating to the metal center [25, 36]. The far-IR region also revealed bands corresponding to ν(Pd–Cl) and ν(Pd–N) vibrations which are absent in the spectra of the free ligands. The FTIR spectra of the complexes are summarized in Tables 1 and 2.

3.5. ^{1}H-NMR Spectra. A comparison of the ^{1}H-NMR spectra of the complexes with those of the free ligands show downfield shifts in the proton signals upon complexation to the palladium(II) ion. As shown in Table 3, the NH_2 cytosine protons of cis-[$PdCl_2$(Gua)(Cyt)] which are directly adjacent to the coordinating N3 ring nitrogen show a downfield shift from 7.06 to 7.70 ppm in the complex. Similarly, the NH_2 protons of guanine observed in the free ligand at 5.57 ppm was shifted to 5.95 ppm showing that these protons

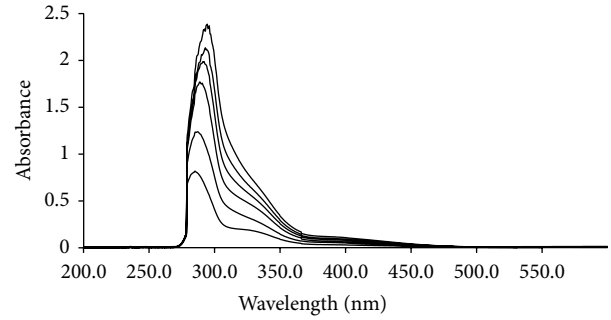

Figure 1: Electronic absorption spectra of cis-[PdCl2(Cyt)(Gua)] showing bathochromism with increasing concentration.

experienced strong deshielding effects upon complexation [37]. Similar chemical shifts were observed for the remaining protons as shown in Table 3. Figure 2 shows the proposed structures of the complexes with labeled atoms.

3.6. Electronic Absorption Spectra, Conductivity, and Magnetic Measurements. The electronic spectra of the complexes were recorded in DMSO (10^{-3} M). This was used to study the effect of complexation on the splitting of the d orbitals in the Pd(II) ion. Significant absorption bands were observed in the region of 280–290, 331–333, 403–407, and 531 nm. These transitions may be attributed to intraligand π-π^* as well as the spin allowed LMCT d-d transitions of

$$\begin{aligned} {}^1A_{1g}\left(d_x{}^2\text{-}d_y{}^2\right) &\longleftarrow {}^1A_{2g}\left(d_{xy}\right), \\ {}^1A_{1g}\left(d_x{}^2\text{-}d_y{}^2\right) &\longleftarrow {}^1E_g\left(d_{xz}, d_{yz}\right), \\ {}^1A_{1g}\left(d_x{}^2\text{-}d_y{}^2\right) &\longleftarrow {}^1B_{1g}\left(d_z{}^2\right), \end{aligned} \tag{2}$$

respectively [37]. These transitions confirm the square planar (D_{4h}) geometry of the Pd(II) ion [31]. Bathochromism was also observed in the spectra of these complexes with increasing concentration as exemplified in Figure 1. This shift in wavelength can be attributed to strong intermolecular hydrogen bonding, a phenomenon which can be likened to the intercalative mode of bonding involving a strong π-π^* stacking interaction between aromatic chromophores and DNA bases [38–40].

The molar conductivities of the complexes measured in DMSO (10^{-3} M) was between 13.58 and 18.48 Ω^{-1} cm^{-2} mol^{-1}

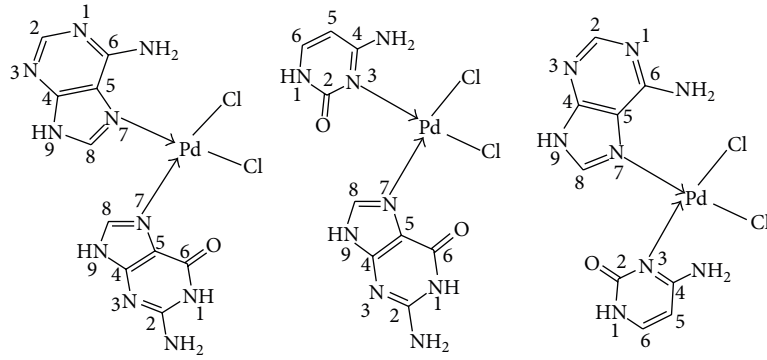

Figure 2: Proposed structures of the palladium(II) chloride complexes.

Table 3: ^{1}H-NMR data of the complexes and the free nucleobase ligands.

Complex	H-NMR[a]
cis-[$PdCl_2$(cyt)(gua)]	5.56 (1H, s, H-5(cyt)), 7.40 (1H, s, H-6(cyt)), 7.70 (s, NH_2(cyt)), 11.27 (1H, s, H-1(cyt)), 5.95 (s, NH_2(gua)), 8.83 (1H, s, H-8(gua)), 9.40 (1H, s, H-9(gua)), 12.03 (1H, s, H-1(gua)).
cis-[$PdCl_2$(cyt)(ad)]	5.90 (1H, s, H-5(cyt)), 7.71 (s, NH_2(cyt)), 8.13 (1H, s, H-6(cyt)), 2.01 (1H, s, H-1(cyt)), 7.70 (s, NH_2(ad)), 8.38 (1H, s, H-2(ad)), 9.37 (1H, s, H-8(ad)), 12.00 (1H, s, H-9(ad))
cis-[$PdCl_2$(ad)(gua)]	8.33 (s, NH_2(ad)), 8.78 (1H, s, H-8(ad)), 9.20 (1H, s, H-2(ad)), 13.45 (1H, s, H-9(ad)), 6.63 (s, NH_2(gua)), 8.29 (1H, s, H-8(gua)), 11.17 (1H, s, H-9(gua)), 14.06 (1H, s, H-1(gua))
Cytosine	5.56 (1H, s, H-5), 7.06 (s, NH_2), 7.32 (1H, s, H-6), 10.56 (1H, s, H-9)
Adenine	7.11 (s, NH_2), 8.10 (2H, s, H-2 and H-8), 12.86 (1H, s, H-9)
Guanine	5.57 (s, NH_2), 7.08 (1H, s, H-8), 10.55 (1H, s, H-9)

[a]25°C in DMSO-d_6 δ [ppm].

indicating the nonelectrolyte nature of these complexes [41–43]. The effective magnetic moment measurement at room temperature confirmed their diamagnetic characters of these complexes thus square-planar geometry.

Based on the above discussion of results and the analytical data obtained from the metal and chloride content analyses we suggest that the structural formulae of the complexes are as given in Figure 2.

3.7. In Vitro Cytotoxicity. The growth inhibition effect of the complexes as well as the free ligands was measured on two leukemia cell lines (human promyelocytic leukemia (HL60) and human histiocytic leukemia (U937)) with curcumin as a positive control. The results as expressed by IC_{50} values (μM) are presented in Table 4. Of the three nucleobase ligands, guanine was the only cytotoxin on HL60 within the working concentration of 0–200 μM. It reported an IC_{50} value of 71.38 ± 9.01 μM. Guanine was also cytotoxic against U937, recording a value of 53.53 ± 8.55 μM.

All the three mixed ligand complexes were significantly cytotoxic with the exception of cis-[$PdCl_2$(Cyt)(Ad)] which did not show any activity on HL60 within the working concentration. From Table 4, cis-[$PdCl_2$(Ad)(Gua)] was the most cytotoxic against both cell lines with IC_{50} values of 11.45 ± 2.91 and 8.31 ± 1.44 μM on the respective cell lines. It was more cytotoxic than the positive control (curcumin) on U937 cell line. The high activity of cis-[$PdCl_2$(Ad)(Gua)] could be attributed to the bulkiness of the nucleobase ligands which has the tendency to significantly reduce the rate of deactivation by sulphur-containing molecules within the cell [44, 45].

Table 4: IC_{50} values of the complexes tested on the respective cell lines.

Complex	Cell line	
	HL60	U937
cis-[$PdCl_2$(Cyt)(Gua)]	132.01 ± 3.44	17.63 ± 1.25
cis-[$PdCl_2$(Cyt)(Ad)]	>200	34.56 ± 2.02
cis-[$PdCl_2$(Ad)(Gua)]	11.45 ± 2.91	8.31 ± 1.44
Adenine	>200	>100
Cytosine	>200	44.95 ± 7.11
Guanine	71.38 ± 9.01	53.53 ± 8.55
Curcumin	5.40 ± 2.55	8.63 ± 1.94

3.8. Antioxidant Activity. Various researchers have used scavenging effect of a chemical on DPPH radical as a quick and reliable parameter to measure the *in vitro* antioxidant activity [12, 13, 46]. This assay is based on the measurement of the decrease in molar absorptivity of DPPH at 517 nm after reaction with the test compound.

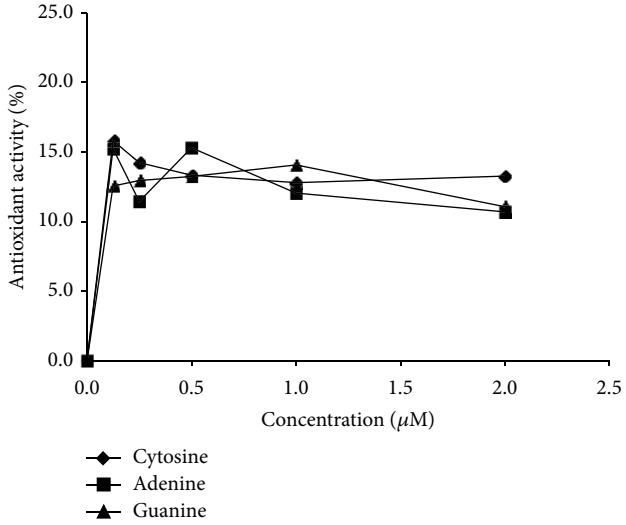

Figure 3: Percent antioxidant activity of the free ligands.

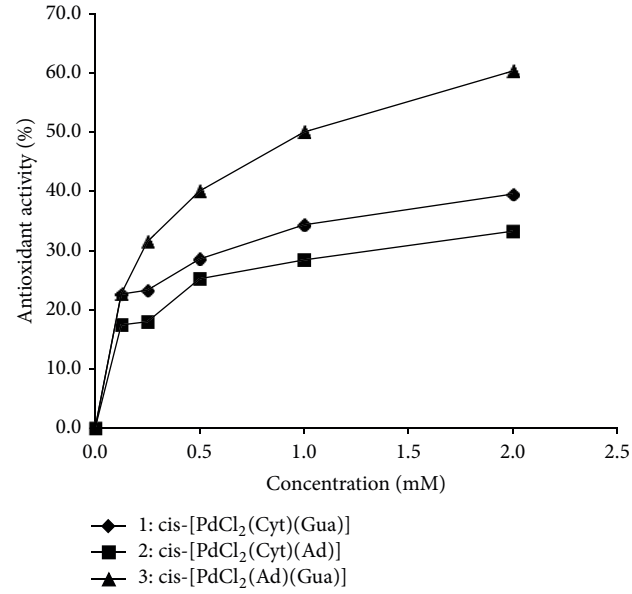

Figure 4: Percent antioxidant activity of the mixed ligand complexes.

From Figure 3, the antioxidant activity of the free ligands was found to be between 10 and 15% but, upon complexation, it increased significantly (Figure 4). The increased antioxidant activity of these complexes can be attributed to the downfield shift of the ^{1}H-NMR signals (Table 3). As reported [12, 13], this phenomenon is as a result of the electron withdrawing effect of the Pd(II) ion which facilitates the release of hydrogen to reduce the DPPH radical. These proton shifts were very profound in cis-$[PdCl_2(Ad)(Gua)]$ with the H-1 proton of guanine absorbing at a chemical shift of 14.06 ppm. This could contribute to the higher activity of cis-$[PdCl_2(Ad)(Gua)]$ with an EC_{50} (concentration of complexes needed to scavenge 50% of the initial DPPH concentration) value of 1.00 mM.

4. Conclusion

In this study, we reported the synthesis and spectroscopic characterization of mixed ligand Pd(II) chloride complexes bearing the nucleobases cytosine, adenine, and guanine. A four-coordinated square-planar geometry was proposed for these complexes based on spectroscopic and magnetic measurements. The ligands and the metal complexes were tested for their antioxidant and *in vitro* cytotoxic activities. All the complexes were significantly cytotoxic with cis-$[PdCl_2(Ad)(Gua)]$ showing higher activity than the positive control (curcumin) on U937 cell line. All the complexes also show significant free radical scavenging ability when tested against DPPH.

Conflict of Interests

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

Acknowledgments

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References

[1] B. T. Khan, S. V. Kumari, and G. N. Goud, "Mixed ligand complexes of platinum (II) with α-amino acids and purines and pyrimidines," *Journal of Coordination Chemistry*, vol. 12, no. 1, pp. 19–25, 1982.

[2] W. Brϋning, E. Freisinger, M. Sabat, R. K. O. Sigél, and B. Lippert, "N1 and N3 linkage Isomers of neutral and protonated cytosine with trans $[(CH_3NH_2)_2Pt(II)]$," *Chemistry*, vol. 8, no. 20, pp. 4681–4692, 2002.

[3] R. Kobetić, D. Gembarovski, A. Višnjevac, B. Žinić, and V. Gabelica-Marković, "ESI-MS studies of palladium (II) complexes with 1-(ρ-toluenesulfonyl) cytosine/cytosinato ligands," *Journal of Mass Spectrometry*, vol. 45, no. 1, pp. 51–64, 2010.

[4] E. G. Bardají, E. Freisinger, B. Costisella et al., "Mixed-metal (platinum, palladium), mixed-pyrimidine (uracil, cytosine) self-assembling metallacalix[n]arenes: dynamic combinatorial chemistry with nucleobases and metal species," *Chemistry*, vol. 13, no. 21, pp. 6019–6039, 2007.

[5] D. Holthenrich, E. Zangrando, E. Chiarparin, B. Lippert, and L. Randaccio, "Cytosine nucleobase as a tridentate ligand: metal binding to N3, N4 and O2 in the trinuclear complex

cis-$[Pt(NH_3)_2(mcyt)_2Pd(en)_2]$-$[NO_3]_4 \cdot H_2O$ (mcyt = 1-methylcytosinate, en = ethane-1,2-diamine)," *Journal of the Chemical Society, Dalton Transactions*, no. 22, pp. 4407–4410, 1997.

[6] C. Navarro-Ranninger, E. I. Montero, I. López-Solera, J. R. Masaguer, and B. Lippert, "A cyclometallated Pd(II) complex containing a cytosine model nucleobase," *Journal of Organometallic Chemistry*, vol. 558, no. 1-2, pp. 103–110, 1998.

[7] A. S. Abu-Surrah, K. A. Abu Safieh, I. M. Ahmad et al., "New palladium(II) complexes bearing pyrazole-based Schiff base ligands: synthesis, characterization and cytotoxicity," *European Journal of Medicinal Chemistry*, vol. 45, no. 2, pp. 471–475, 2010.

[8] A. Pañella, J. Pons, J. García-Antón, X. Solans, M. Font-Bardia, and J. Ros, "Synthesis of new palladium(II) complexes containing hemilabile N-(alkylamino)pyrazole ligands: spectroscopic analysis and crystal structure of $PdCl(ddae)Cl \cdot H_2O$ ddae = Bis[2-(3,5-dimethyl-1-pyrazolyl)ethyl]ethylamine," *European Journal of Inorganic Chemistry*, no. 8, pp. 1678–1685, 2006.

[9] S. Pérez-Yáñez, O. Castillo, J. Capeda, J. P. Garcia-Teran, A. Luque, and P. Roman, "Supramolecular architectures of metal-oxalato complexes containing purine nucleobases," *Inorganica Chimica Acta*, 2010.

[10] G. Cervantes, J. J. Fiol, A. Terrón et al., "Synthesis and characterization of nickel(II) complexes of purine and pyrimidine bases. Crystal and molecular structure of trans-bis(cytosine-O^2)bis(ethylenediamine)nickel(II) bis(tetraphenylborate). An unusual metal binding mode of cytosine," *Inorganic Chemistry*, vol. 29, no. 26, pp. 5168–5173, 1990.

[11] X. Zhong, W. Slough, R. Pandey, and C. Friedrich, "Interaction of nucleobases with silicon nanowires: a first principle study," *Chemical Physics Letters*, vol. 553, pp. 55–58, 2012.

[12] A. A. Al-Amiery, A. A. H. Kadhum, and A. B. Mohammed, "Antifungal and antioxidant activities of pyrolidone thiosemicarbazone complexes," *Bioinorganic Chemistry and Applications*, vol. 2012, Article ID 795812, 6 pages, 2012.

[13] A. Choudhary, R. Sharma, M. Nagar, M. Mohsin, and H. S. Meena, "Synthesis, characterization and antioxidant activity of some transition metal complexes with terpenoid derivatives," *Journal of the Chilean Chemical Society*, vol. 56, no. 4, pp. 911–917, 2011.

[14] A. S. Mildvan and M. Cohn, "Kinetic and magnetic resonance studies of the pyruvate kinase reaction. II. Complexes of enzyme, metal, and substrates," *Journal of Biological Chemistry*, vol. 241, no. 5, pp. 1178–1193, 1966.

[15] G. M. R. Tombo and D. Bellus, "Chiralität und pflanzenschutz," *Angewandte Chemie*, vol. 103, no. 10, pp. 1219–1241, 1991.

[16] A. A. El-Sherif, M. R. Shehata, M. M. Shoukry, and M. H. Barakat, "Thermodynamic investigation and mixed-ligand complex formation of 1,4-bis-(3-aminopropyl)-piperazine and biorelevant ligands," *Bioinorganic Chemistry and Applications*, vol. 2012, Article ID 984291, 10 pages, 2012.

[17] A. M. Ryzhakov, M. S. Gruzdev, D. F. Pyreu, E. V. Kozlovskii, and R. S. Kumeev, "Thermodynamics of mixed-ligand complexation of mercury(II) ethylenediaminetetraacetate with histidine and lysine in aqueous solution," *Russian Journal of Coordination Chemistry*, vol. 36, no. 8, pp. 565–571, 2010.

[18] R. Ahuja and K. Dwivedi, "Thermodynamic equilibrium constants of mixed-ligand complexes of Cu(II) with ethyleneglycol-bis(2-aminoethylether)-N,N,N′,N′ tetraacetic acid as primary and alanine and phenylalanine as secondary ligands," *Asian Journal of Chemistry*, vol. 5, no. 1, pp. 54–59, 1993.

[19] D. B. Berezin, O. V. Shukhto, and P. A. Shatunov, "Effect of ligand nonplanarity and solvent nature on the kinetic stability of zinc porphyrin complexes," *Russian Journal of General Chemistry*, vol. 78, no. 5, pp. 997–1004, 2008.

[20] G. Rajendran and C. R. K. Kumari, "Kinetic and antimicrobial studies on some novel mixed ligand complexes of thorium(IV) and dioxouranium(VI) ions," *Asian Journal of Chemistry*, vol. 16, no. 2, pp. 643–648, 2004.

[21] J. Nemes, I. Tóth, and L. Zékány, "Formation kinetics of an aluminium(III)-ethylenedinitrilotetraacetate-fluoride mixed ligand complex," *Journal of the Chemical Society, Dalton Transactions*, no. 16, pp. 2707–2713, 1998.

[22] K. Rai and F. Khan, "A study of stability constants of [Zn-L-aminoacidate-vitamin-PP] systems," *American Journal of Chemistry*, vol. 2, no. 6, pp. 299–305, 2012.

[23] J. Reedijk, "Metal-ligand exchange kinetics in platinum and ruthenium complexes: significance for effectiveness as anticancer drugs," *Platinum Metals Review*, vol. 52, no. 1, pp. 2–11, 2008.

[24] Y. Zhang, X. Wang, W. Fang et al., "Synthesis and *in vitro* antitumor activity of two mixed-ligand oxovanadium(IV) complexes of schiff base and phenanthroline," *Bioinorganic Chemistry and Applications*, vol. 2013, Article ID 437134, 14 pages, 2013.

[25] Y. Prashanthi, K. Kiranmai, Ira, K. Sathish Kumar, V. K. Chityala, and Shivaraj, "Spectroscopic characterization and biological activity of mixed-ligand complexes of Ni(II) with 1,10-phenanthroline and heterocyclic schiff bases," *Bioinorganic Chemistry and Applications*, vol. 2012, Article ID 948534, 8 pages, 2012.

[26] H. Khan, A. Badshah, M. Said et al., "Anticancer metallopharmaceutical agents based on mixed-ligand palladium(II) complexes with dithiocarbamates and tertiary organophosphine ligands," *Applied Organometallic Chemistry*, vol. 27, no. 7, pp. 387–395, 2013.

[27] G. D. Geromichalos, G. A. Katsoulos, D. T. Trafalis, C. C. Hadjikostas, and A. Papageorgiou, "Synergistic interaction between a novel mixed ligand copper(II) chelate complex and a panel of anticancer agents in T47D human breast cancer cells *in vitro*," *Journal of B.U.ON*, vol. 10, no. 2, pp. 235–240, 2005.

[28] M. Paula, M. Marques, R. Valero, S. F. Parker, J. Tomkinson, and L. A. E. Batista de Carvalho, "Polymorphism in cisplatin anticancer drug," *The Journal of Physical Chemistry B*, vol. 117, no. 21, pp. 6421–6429, 2013.

[29] K. Pi-Chang and F. D. Rochon, "Palladium compounds of pyrimidine and pyridine derivatives—proposed structure assignment changes," *Canadian Journal of Chemistry*, vol. 59, no. 23, pp. 3293–3296, 1981.

[30] A. M. A. Khader and K. S. Prasad, "Extractive spectrophotometry of palladium(II) with 3,4,5-trimethoxybenzaldehyde thiosemicarbazone," *Turkish Journal of Chemistry*, vol. 20, no. 3, pp. 222–227, 1996.

[31] G. L. Meissler and D. A. Tarr, *Inorganic Chemistry*, Pearson Education Inc, Upper Saddle River, NJ, USA, 3rd edition, 2004.

[32] D. P. Shoemaker, C. W. Garland, J. I. Steinfeld, and J. W. Nibler, *Experiments in Physical Chemistry*, McGraw-Hill, New York, NY, USA, 4th edition, 1981.

[33] W. Gao, S. Bussom, S. P. Grill et al., "Structure-activity studies of phenanthroindolizidine alkaloids as potential antitumor agents," *Bioorganic and Medicinal Chemistry Letters*, vol. 17, no. 15, pp. 4338–4342, 2007.

[34] M. S. Blois, "Antioxidant determinations by the use of a stable free radical," *Nature*, vol. 181, no. 4617, pp. 1199–1200, 1958.

[35] A. I. Matesanz, J. M. Pérez, P. Navarro, J. M. Moreno, E. Colacio, and P. Souza, "Synthesis and characterization of novel palladium(II) complexes of bis(thiosemicarbazone). Structure, cytotoxic activity and DNA binding of Pd(II)-benzyl bis(thiosemicarbazonate)," *Journal of Inorganic Biochemistry*, vol. 76, no. 1, pp. 29–37, 1999.

[36] P. A. Ajibade and O. G. Idemudia, "Synthesis, characterization and antibacterial studies of Pd(II) and Pt(II) complexes of some diaminopyrimidine derivatives," *Bioinorganic Chemistry and Applications*, vol. 2013, Article ID 549549, 8 pages, 2013.

[37] E. Szlyk, A. Grodzicki, L. Pazderski et al., "Palladium(II) chloride complexes with 1, 2, 4-triazolo[1, 5-a]-pyrimidines: x-ray, ^{15}N-^{1}H NMR and ^{15}N CP MAS studies," *Journal of the Chemical Society, Dalton Transactions*, no. 6, pp. 867–872, 2000.

[38] J. Liu, T. Zhang, T. Lu et al., "DNA-binding and cleavage studies of macrocyclic copper(II) complexes," *Journal of Inorganic Biochemistry*, vol. 91, no. 1, pp. 269–276, 2002.

[39] H. Wu, F. Kou, F. Jai, B. Liu, J. Yuan, and Y. Bai, "Synthesis and Characterization of the ligand based on benzimidazole and its copper complex: DNA-binding and antioxidant activity," *Bioinorganic Chemistry and Applications*, vol. 2011, Article ID 105431, 11 pages, 2011.

[40] G. Aragay, J. Pons, J. García-Antón et al., "Synthesis and Characterization of New Palladium(II) Complexes Containing N-Alkylamino-3,5-diphenylpyrazole Ligands. Crystal Structure of [PdCl(L2)](BF4) L2 = bis[2-(3,5-diphenyl-1-pyrazolyl)ethylamine," *Australian Journal of Chemistry*, vol. 62, no. 5, pp. 475–482, 2009.

[41] W. J. Geary, "The use of conductivity measurements in organic solvents for the characterisation of coordination compounds," *Coordination Chemistry Reviews*, vol. 7, no. 1, pp. 81–122, 1971.

[42] Ü. Ö. Özmen and G. Olgun, "Synthesis, characterization and antibacterial activity of new sulfonyl hydrazone derivatives and their nickel(II) complexes," *Spectrochimica Acta A*, vol. 70, no. 3, pp. 641–645, 2008.

[43] C. M. Sharaby, "Synthesis, spectroscopic, thermal and antimicrobial studies of some novel metal complexes of Schiff base derived from [N1-(4-methoxy-1,2,5-thiadiazol-3-yl)sulfanilamide] and 2-thiophene carboxaldehyde," *Spectrochimica Acta A*, vol. 66, no. 4-5, pp. 1271–1278, 2007.

[44] K. A. Gelmon, D. Stewart, K. N. Chi et al., "A phase I study of AMD473 and docetaxel given once every 3 weeks in patients with advanced refractory cancer: a National Cancer Institute of Canada-Clinical Trials Group trial, IND 131," *Annals of Oncology*, vol. 15, no. 7, pp. 1115–1122, 2004.

[45] L. Kelland, "Broadening the clinical use of platinum drug-based chemotherapy with new analogues: satraplatin and picoplatin," *Expert Opinion on Investigational Drugs*, vol. 16, no. 7, pp. 1009–1021, 2007.

[46] A. A. H. Kadhum, A. B. Mohamad, A. A. Al-Amiery, and M. S. Takriff, "Antimicrobial and antioxidant activities of new metal complexes derived from 3-aminocoumarin," *Molecules*, vol. 16, no. 8, pp. 6969–6984, 2011.