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Stereospecific ligands and their complexes. XXII. Synthesis and antitumor activity of palladium(II) complexes with some esters of (*S,S*)-ethylenediamine-*N,N′*-di-(2,2′-di(4-hydroxy-benzyl))-acetic acid ☆

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ABSTRACT

Four new ligands and their palladium(II) complexes of general formula R_2-*S,S*-eddtyr (**L1**–**L4**) and [$PdCl_2$(R_2-*S,S*-eddtyr)] (**C1**–**C4**) (R = ethyl, *n*-propyl, *n*-butyl and *n*-pentyl; *S,S*-eddtyr·2HCl = ethylenediamine-*N,N′*-di-(2,2′-di(4-hydroxy-benzyl))-acetic acid dihydrochloride have been synthesized and characterized by microanalysis, infrared, ^{1}H and ^{13}C NMR spectroscopy. Cytotoxicity for ligands and complexes on two different cell lines (human breast cancer, MDA-MB-231 and human lung cancer, A549 cell lines) and human chronic lymphocytic leukemia cells (CLL) was investigated using MTT (3-[4,5-dimethylthiazol-2-yl]-2,5-diphenyltetrazolium bromide) assay.

1. Introduction

The discovery of cisplatin led scientists to synthesize many platinum-based drugs that could potentially be less toxic to healthy tissue [1–3] and overcome the resistance of some tumors to this drug [4,5].

The success of metallodrugs is closely linked to the proper choice of ligands, as they play a crucial role in modifying reactivity and lipophilicity, in stabilizing specific oxidation states and in imparting substitution inertness [6,7].

The aim of developing non-platinum anticancer complexes is overcoming the main limits of platinum drugs: narrow range of activity, acquired after treatment resistance, and severe toxicity on healthy tissue [1–5]. Non-platinum complexes may exhibit anticancer activity and toxic side-effects markedly different from that of platinum based drugs for a number of reasons. Palladium derivatives have also been a subject of wide examination due to their structural analogy with Pt(II) complexes. However, initial results were not very encouraging because the Pd(II) derivatives generally showed lower anticancer activity than cisplatin. This behavior may be related to the more labile nature of palladium(II) relative to platinum(II) complexes [8,9]. Rapid ligand exchange was thought to diminish the possibility of Pd(II) complexes to reach the biological target unchanged and increase the risk of adverse effects on biochemical processes occurring in normal cells. In order to overcome these problems, several authors [10] have suggested that the use of chelating ligands may reduce the reactivity of the palladium center. Generally, Pd(II) complexes are divided into two major groups: Pd(II) complexes with sulfur donor ligands and Pd(II) complexes with nitrogen and other donor ligands. Cytotoxic activity of Pd(II) complexes with thiosemicarbazones was shown against MCF-7, TK-10 and UACC-62 human tumor cell lines. They showed lower IC_{50} values and were more cytotoxic in these cell lines than cisplatin [11]. Other Pd(II) complexes with sulfur donor ligands showed a moderate to low cytotoxicity *in vitro* against various human tumor cell lines [12]. Some Pd(II) complexes containing the bidentate ligand 9-aminoacridine showed higher cytotoxic activity against human ovarian cancer cells than both cisplatin and the free ligands [13].

A number of palladium(II) complexes with neutral ligands such as pyridine derivatives [14,15], phosphonate derivatives of quinoline [16] or pyrazole derivatives [17] have been investigated and their significant cytotoxic activity has been proved. *In vitro* antitumor activity of some complexes was compared to that of cisplatin [18–22], and as a results of these findings *N,N* bidentate esters, R_2edda-type ligands, containing hiral C-atoms with *S*

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absolute configuration originating from natural aminoacids are now mostly used in our studies. Recently, we have reported the synthesis and characterization of the palladium(II) complexes with dialkyl esters of [(*S,S*)-H_2-R_2eddip]Cl_2 [23], H_2-*S,S*-eddp [24], H_2-(*S,S*)-eddv [25], H_2-(*S,S*)-eddba [26], (R = ethyl, *n*-propyl, *n*-butyl and *n*-pentyl) and their antitumor and antimicrobial activities. As continuation of our work we report herein the synthesis, characterization and cytotoxicity of four novel R_2edda-type ligand precursors: *O,O′*-diethyl (**L1**), *O,O′*-dipropyl (**L2**), *O,O′*-dibutyl (**L3**), *O,O′*-dipentyl-(*S,S*)-ethylenediamine-*N,N′*-di-(2,2′-di(4-hydroxy-benzyl))-acetic acid dihydrochlorides dihydrates (**L4**) and their corresponding palladium(II) complexes: dichlorido-(*O,O′*-diethyl-(*S,S*)-ethylenediamine-*N,N′*-di-(2,2′-di(4-hydroxy-benzyl))-acetate)-palladium(II) (**C1**), dichlorido-(*O,O′*-dipropyl-(*S,S*)-ethylenediamine-*N,N′*-di-(2,2′-di(4-hydroxy-benzyl))-acetate)-palladium(II) (**C2**), dichlorido-(*O,O′*-dibutyl-(*S,S*)-ethylenediamine-*N,N′*-di-(2,2′-di(4-hydroxy-benzyl))-acetate)-palladium(II) (**C3**), dichlorido-(*O,O′*-dipentyl-(*S,S*)-ethylenediamine-*N,N′*-di-(2,2′-di(4-hydroxy-benzyl))-acetate)-palladium(II) (**C4**).

2. Experimental

2.1. Chemistry

2.1.1. Reagents and instruments

(*S,S*)-ethylenediamine-*N,N′*-di-(2,2′-di(4-hydroxy-benzyl))-acetic acid dihydrochloride [(H_2-(*S,S*)-eddtyr)] Cl_2 was prepared using similar methods described earlier [27]. $K_2[PdCl_4]$ was purchased from Merck and used without further purification. Alcohols were dried by standard methods. Infrared spectra were recorded by Perkin-Elmer Spectrum One FT-IR spectrometer using the KBr pellet technique (4000–400 cm^{-1}). ^1H and ^{13}C NMR (Fig. 1) spectra were recorded by Varian Gemini-2000 (200 MHz) spectrometer in DMSO-d_6 (ligands and complexes) using tetramethylsilane as internal standard.

Elemental microanalyses for C, H and N were performed by standard methods by Vario EL *III* C, H, N elemental analyzer.

2.1.2. Preparation of *O,O′*-dialkyl esters of (*S,S*)-ethylenediamine-*N,N′*-di-(2,2′-di(4-hydroxy-benzyl))-acetic acid dihydrochloride, R_2-*S,S*-eddtyr·2HCl

In 40 mL of dry alcohol (ethanol, 1-propanol, 1-butanol or 1-pentanol) saturated with gas HCl, 1.50 g (3.25 mmol) of H_2-(*S,S*)-eddtyr was added and the mixture was refluxed for 12 h (Scheme 1). The mixture was filtered off and the filtrate was left for a few days in the refrigerator. The esters were recrystallized from hot alcohol used for each reaction.

O,O′-diethyl-(*S,S*)-ethylenediamine-*N,N′*-di-(2,2′-di(4-hydroxy-benzyl))-acetic acid dihydrochloride dihydrate, det-*S,S*-eddtyr·2HCl·$2H_2O$ (**L1**). Yield: 0.87 g (48.31%). Anal. calc. for $C_{24}H_{38}Cl_2N_2O_8$ (M_r = 553.464): C, 52.08; H, 6.92; N, 5.06. Found: C, 51.89; H, 6.73; N, 4.94%.

^1H NMR (200 MHz, DMSO, δ ppm): 3.01 (m, 4H, C^1H_2), 3.29 (t, 2H, C^2H), 1.12 (d, 4H, C^3H_2), 7.02 (AB q, 4H, $C^{5,9}H$), 6.73 (AB q, 4H, $C^{6,8}H$), 4.13 (q, 4H, $C^{11}H_2$), 0.85 (t, 6H, $C^{12}H_3$), ^{13}C NMR (200 MHz, DMSO, δ ppm): 53.58 (C^1H_2), 62.03 (C^2H), 27.83 (C^3H_2), 130.60 (C^4), 156.89 ($C^{5,9}H$), 115.52 ($C^{6,8}H$), 168.29 (C^7), 170.44 (C^{10}), 60.66 ($C^{11}H_2$), 14.06 ($C^{12}H_3$). IR (cm^{-1}): 3428, 2925, 2853, 1737, 1638, 1614, 1516, 1445, 1400, 1384, 1229, 1106, 1058, 826, 732, 519.

O,O′-dipropyl-(*S,S*)-ethylenediamine-*N,N′*-di-(2,2′-di(4-hydroxy-benzyl))-acetic acid dihydrochloride dihydrate, dpr-*S,S*-eddtyr·2HCl·$2H_2O$ (**L2**). Yield: 0.88 g (46,42%). Anal. calc. for $C_{26}H_{42}Cl_2N_2O_8$ (M_r = 581.516): C, 53.69; H, 7.28; N, 4.82. Found: C, 53.44; H, 7.03; N, 4.85%.

^1H NMR (200 MHz, DMSO, δ ppm): 2.99 (m, 4H, C^1H_2), 3.37 (t, 2H, C^2H), 1.49 (d, 4H, C^3H_2), 6.99 (AB q, 4H, $C^{5,9}H$), 6.69 (AB q, 4H, $C^{6,8}H$), 4.01 (t, 4H, $C^{11}H_2$), 0.82 (m, 4H, $C^{12}H_2$), 0.72 (t, 6H, $C^{13}H_3$), ^{13}C NMR (200 MHz, DMSO, δ ppm): 53.60 (C^1H_2), 60.68 (C^2H), 35.39 (C^3H_2), 124.51 (C^4), 130.55 ($C^{5,9}H$), 115.52 ($C^{6,8}H$), 156.87 (C^7), 169.17 (C^{10}), 67.12 ($C^{11}H_2$), 21.36 ($C^{12}H_2$), 10.19 ($C^{13}H_3$). IR (cm^{-1}): 3419, 2925, 2853, 1734, 1615, 1516, 1444, 1400, 1384, 1227, 1107, 1056, 824, 730, 519.

O,O′-dibutyl-(*S,S*)-ethylenediamine-*N,N′*-di-(2,2′-di(4-hydroxy-benzyl))-acetic acid dihydrochloride dihydrate, dbu-*S,S*-eddtyr·2HCl·$2H_2O$ (**L3**). Yield: 0.87 g (44.13%). Anal. calc. for $C_{28}H_{46}Cl_2N_2O_8$ (M_r = 609.568): C, 55.17; H, 7.61; N, 4.59. Found: C, 54.97; H, 7.43; N, 4.76%.

^1H NMR (200 MHz, DMSO, δ ppm): 2.94 (m, 4H, C^1H_2), 3.01 (t, 2H, C^2H), 1.46 (d, 4H, C^3H_2), 7.03 (AB q, 4H, $C^{5,9}H$), 6.73 (AB q, 4H, $C^{6,8}H$), 4.04 (t, 4H, $C^{11}H_2$), 1.22 (m, 4H, $C^{12}H_2$), 1.16 (m, 4H, $C^{13}H_2$), 0.83

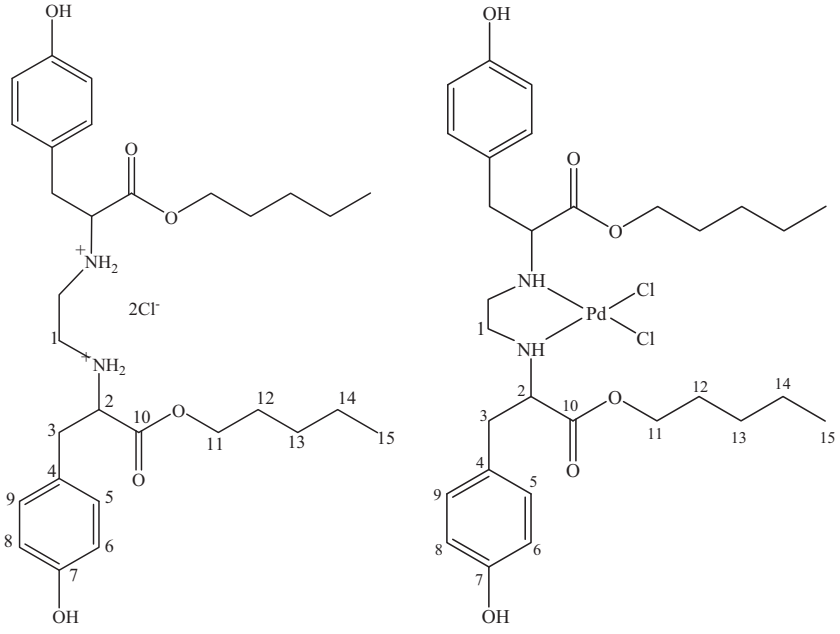

Fig. 1. Numbering of compounds used for NMR data.