Available online at www.sciencedirect.com
SCIENCE DIRECT®
Inorganica Chimica Acta 358 (2005) 2239–2245
Inorganica Chimica Acta
www.elsevier.com/locate/ica

Syntheses and activity of some platinum(IV) complexes with *N*-methyl derivate of glycine and halogeno ligands against HeLa, K562 cell lines and human PBMC

Tibor J. Sabo [a], Vesna M. Đinović [a], Goran N. Kaluđerović [a,*], Tatjana P. Stanojković [b], Goran A. Bogdanović [c], Zorica D. Juranić [b]

[a] *Faculty of Chemistry, University of Belgrade, Studentski trg 12-16, P.O. Box 158, 11001 Belgrade, Serbia and Montenegro*
[b] *Institute of Oncology and Radiology, 11000 Belgrade, Serbia and Montenegro*
[c] *VINČA Institute of Nuclear Sciences, P.O. Box 522, Belgrade 11001, Serbia and Montenegro*

Received 11 July 2004; accepted 11 January 2005

Abstract

Four platinum(IV) complexes, *trans*,*trans*-dichlorobis(*N*,*N*-dimethylglycinato)platinum(IV), *trans*,*trans*-[Pt(dmgly)$_2$Cl$_2$] (**1**) and *trans*,*trans*-dibromobis(*N*,*N*-dimethylglycinato)platinum(IV), *trans*,*trans*-[Pt(dmgly)$_2$Br$_2$] (**2**), as well as, *trans*,*trans*-dichlorobis(*N*-methylglycinato)platinum(IV), *trans*,*trans*-[Pt(sar)$_2$Cl$_2$] (**3**) and *trans*,*trans*-dibromobis(*N*-methylglycinato)platinum(IV), *trans*,-*trans*-[Pt(sar)$_2$Br$_2$] (**4**) (with configuration index for all complexes *OC*-6–14), were synthesized and characterized by elemental analysis, infrared and ^{1}H NMR spectroscopy. In the aim to assess the selectivity in the antitumor action of these complexes, the antiproliferative action of these compounds was determined to human adenocarcinoma HeLa cells; to human myelogenous leukemia K562 cells and to normal immunocompetent cells; i.e., on human PBMC. The details of the crystal structure synthesized *trans*,*trans*-[Pt(sar)$_2$Br$_2$] complex were also reported here. In the crystal structure of *trans*,*trans*-[Pt(sar)$_2$Br$_2$], the Pt(IV) ion had a deformed octahedral coordination with both *N*-methylglycinates and bromides bonded *trans* to one another and with the N–Pt–Br bond angles of 84.1(4) and 95.9(4)°. The *trans*,*trans*-[Pt(sar)$_2$Br$_2$] complex molecules form 2D-layers with multiple N–H···O and C–H···O hydrogen bonds.

Keywords: Platinum (IV) complexes; Bidentate ligands; Sarcosine; Crystal structure; Neoplastic cells; PBMC

1. Introduction

The observation that neutral platinum coordination compounds inhibit division and cell growth has generated much interest in the potential value of inorganic drugs in the field of cancer chemotherapy [1–6]. Cisplatin has been shown to have potent antitumor activity and is nowadays routinely employed in the treatment of several cancers [7,8]. However, because of its severe side-effect a need for new platinum complexes has arisen, and several new derivatives have been synthesized and tested against various tumor model systems, with the hope of discovering new drugs with improved properties [5]. In the most successful second-generation cisplatin analogs (i.e., carboplatin), the chloride ligands have been replaced by a carboxylate. Therefore, there has been an increasing interest in platinum(IV) complexes with biologically important ligands, such as aminocarboxylate ligands [9], because of recent development of new Pt(IV)-containing anti-cancer drugs and the evidence that they may react with the DNA without being reduced to Pt(II) [1,2].

* Corresponding author. Tel.: +3811 3282 111/736; fax: +38111 638 785.
E-mail address: goran@chem.bg.ac.yu (G.N. Kaluđerović).

0020-1693/$ - see front matter © 2005 Elsevier B.V. All rights reserved.
doi:10.1016/j.ica.2005.01.007

This paper reports on the synthesis of novel platinum(IV) complexes with *N*-methyl derivate of glycine and halogeno ligands by simple ligand exchange reactions with $K_2[PtX_6]$, (where X is a Cl^- or Br^-), as starting materials, in the presence of lithium hydroxide.

2. Experimental

2.1. Starting materials and physical methods

Potassium hexachloroplatinate(IV), potassium hexabromoplatinate(IV), *N*,*N*-dimethylglycine (dmgly) and *N*-methylglycine (sar) were obtained by Merck and used without further purification.

For infrared spectra, a Perkin–Elmer FTIR 31725-X spectrophotometer and KBr pellet technique were employed. ^{1}H NMR spectra were recorded on a Varian Gemini-200 NMR spectrometer using TMS in DMSO as internal reference. Elemental analyses were done on a Vario III CHNOS Elemental Analyzer, Elemental Analysensysteme GmbH.

2.2. Preparation of platinum complexes

2.2.1. Preparation of trans, trans-dichlorobis(N,N-dimethylglycinato)platinum(IV), trans,trans-[Pt(dmgly)$_2$Cl$_2$] (1)

$K_2[PtCl_6]$ (0.129 g, 0.266 mmol) was dissolved in 10.0 cm^3 water on a steam bath and *N*,*N*-dimethylglycine (0.071 g, 0.798 mmol) was added. The mixture was stirred for 12 h and during this period 10.0 cm^3 LiOH (0.019 g, 0.795 mmol) was introduced. The complex **1**, as an orange precipitate, was separated by filtration, washed with water and air-dried. Yield: 0.076 g (61%). *Anal*. Calc. for **1** *trans*,*trans*-[Pt(dmgly)$_2$Cl$_2$]=$C_8H_{16}Cl_2N_2O_4Pt$ (*Mr* = 470.1): C, 20.43; H, 3.40; N, 5.96. Found: C, 20.70; H, 3.44; N, 5.96%. ^{1}H NMR: δ (ppm, DMSO-d_6) 3.4–4.4 (q, CH_2), 2.8 and 2.9 (s, CH_3).

2.2.2. Preparation of trans, trans-dibromobis(N,N-dimethylglycinato)platinum(IV), trans,trans-[Pt(dmgly)$_2$Br$_2$] (2)

The complex **2** was prepared as described for complex **1**, using $K_2[PtBr_6]$ (0.200 g, 0.266 mmol) instead of $K_2[PtCl_6]$. Yield: 0.087 g (59%). *Anal*. Calc. for **2** *trans*, *trans*-[Pt(dmgly)$_2$Br$_2$]=$C_8H_{16}Br_2N_2O_4Pt$ (*Mr* = 558.90): C, 17.18; H, 2.86; N, 5.01. Found: C, 17.23; H, 3.01; N, 5.10%. ^{1}H NMR: δ (ppm, DMSO-d_6) 3.4–4.4 (q, CH_2), 2.7 and 2.8 (s, CH_3).

2.2.3. Preparation of trans,trans-dichlorobis(N-methylglycinato)platinum(IV), trans,trans-[Pt(sar)$_2$Cl$_2$] (3)

$K_2[PtCl_6]$ (0.129 g; 0.266 mmol) was dissolved in 10.0 cm^{-3} water on a steam bath and sarcosine (0.071 g, 0.798 mmol) was added. The mixture was stirred for 12 h and during this period 10.0 cm^{-3} LiOH (0.019 g, 0.795 mmol) was introduced. The complex **3**, as an orange precipitate, was separated by filtration, washed with water and air-dried. Yield: 0.085 g (60%). *Anal*. Calc. for **3** *trans*,*trans*-[Pt(sar)$_2$Cl$_2$]=$C_6H_{12}Cl_2N_2O_4Pt$ (*Mr* = 442.08): C, 16.29; H, 2.71; N, 6.33. Found: C, 16.37; H, 2.70; N, 6.40%. ^{1}H NMR: δ (ppm, DMSO-d_6) 3.7–4.2 (q, CH_2), 2.8 and 2.9 (s, CH_3).

2.2.4. Preparation of trans, trans-dibromobis(N-methylglycinato)platinum(IV), trans, trans-[Pt(sar)$_2$Br$_2$] (4)

The complex **4** was prepared as described for complex **3**, using $K_2[PtBr_6]$ (0.200 g, 0.266 mmol) instead of $K_2[PtCl_6]$. Yield: 0.065 g (56%). *Anal*. Calc. for **4** *trans*, *trans*-[Pt(sar)$_2$Br$_2$]=$C_6H_{12}Br_2N_2O_4Pt$ (*Mr* = 530.90): C, 13.56; H, 2.26; N, 5.27. Found: C, 13.85; H, 2.48; N, 5.26%. ^{1}H NMR: δ (ppm, DMSO-d_6) 3.7–4.2 (q, CH_2), 2.7 and 2.9 (s, CH_3).

2.3. In vitro cytotoxicity studies

Stock solutions of investigated platinum complexes were made in dimethyl sulfoxide (DMSO) at concentration of 10 mM, or 20 mM, filtered through Millipore filter, 0.22 μm, before use, or were resuspended in nutrient medium to homogenous suspension, in concentration of 600 μM, and diluted by nutrient medium to various working concentrations. Nutrient medium was RPMI 1640 medium, without phenol red, supplemented with L-glutamine (3 mM), streptomycin (100 μg/ml), and penicillin (100 IU/ml), 10% heat inactivated (56 °C) fetal bovine serum (FBS) and 25 mM Hepes, and was adjusted to pH 7.2 by bicarbonate solution. MTT, 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyl tetrazolium bromide, was dissolved (5 mg/ml) in phosphate buffer saline, pH 7.2, and filtered through Millipore filter, 0.22 μm, before use. All reagents were Sigma products.

2.3.1. Cell culture

Human cervix adenocarcinoma HeLa cells were cultured as monolayers in the nutrient medium, while human myelogenous leukemia K562 cells, were maintained as suspension culture. The cells were grown at 37 °C in 5% CO_2 and humidified air atmosphere.

2.3.2. Treatment of cell lines

HeLa cells were seeded (2000 cells per well) into 96-well microtiter plates and 20 h later, after the cell adherence, five different concentrations of investigated compounds (from 12.5 to 200 μM) were added to the wells. Only nutrient medium was added to the cells in the control wells. Investigated compounds were added to suspension of leukemia K562 cells (3000 cells per well) 2 h after the cell seeding, in the same final concen-