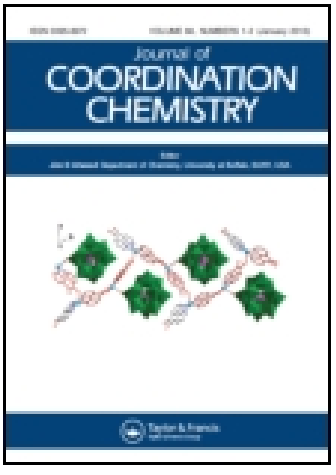


Journal of Coordination Chemistry
Publication details, including instructions for authors and subscription information:
http://www.tandfonline.com/loi/gcoo20

Calixplatin: novel potential anticancer agent based on the platinum complex with functionalized calixarene

Fazel Nasuhi Pur[ab] & Karim Akbari Dilmaghani[a]

[a] Faculty of Science, Department of Chemistry, Urmia University, Urmia, Iran

[b] Health Technology Incubator Center, Urmia University of Medical Science, Urmia, Iran

Accepted author version posted online: 05 Feb 2014.Published online: 05 Mar 2014.

To cite this article: Fazel Nasuhi Pur & Karim Akbari Dilmaghani (2014) Calixplatin: novel potential anticancer agent based on the platinum complex with functionalized calixarene, Journal of Coordination Chemistry, 67:3, 440-448, DOI: 10.1080/00958972.2014.890718

To link to this article: http://dx.doi.org/10.1080/00958972.2014.890718

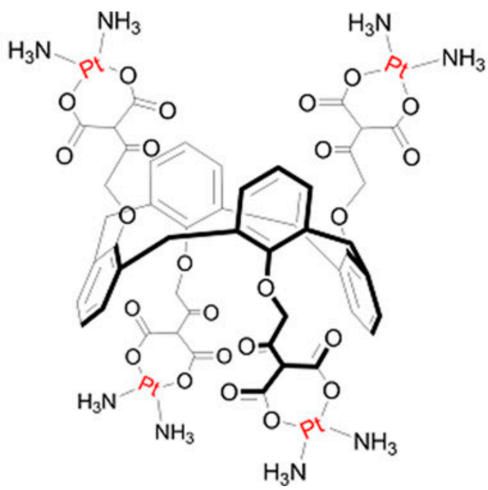
Journal of Coordination Chemistry, 2014
Vol. 67, No. 3, 440–448, http://dx.doi.org/10.1080/00958972.2014.890718

Calixplatin: novel potential anticancer agent based on the platinum complex with functionalized calixarene

FAZEL NASUHI PUR*†‡ and KARIM AKBARI DILMAGHANI*†

†Faculty of Science, Department of Chemistry, Urmia University, Urmia, Iran
‡Health Technology Incubator Center, Urmia University of Medical Science, Urmia, Iran

(*Received 29 September 2013; accepted 10 January 2014*)



A functionalized calixarene with four platinum(II) centers was synthesized as a potential anticancer agent. The colorimetric cytotoxicity tests showed that it has amplified activity with respect to carboplatin against three different human carcinoma (non-small-cell lung, hepatocellular, and breast cancer) cell lines. It was most cytotoxic against the lung cancer cell line ($IC_{50} = 2.6$ µM).

Keywords: Synthesis; Agent; Platinum complex; Calixarene; Carboplatin

1. Introduction

Cisplatin (platinol, **A**), carboplatin (paraplatin, **B**), and oxaliplatin (eloxatine, **C**) are anticancer drugs (figure 1) which are efficiently used in clinics all over the world.

*Corresponding authors. Email: fazelnasuhi@gmail.com (F.N. Pur); k.adilmaghani@urmia.ac.ir (K.A. Dilmaghani)

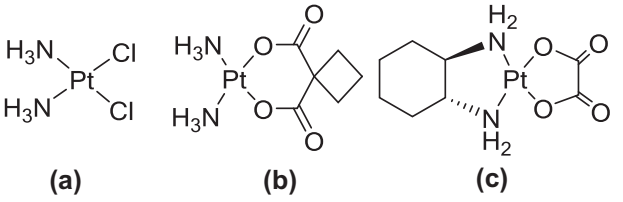


Figure 1. Cisplatin (a), carboplatin (b), and oxaliplatin (c).

Platinum-based therapy is accompanied by severe toxic side effects. The design of platinum complexes equipped with selectivity for tumoral tissue and exhibiting a lack of systemic toxicity is a great hope in the fight against cancer, and also an impetus to the expanding field of bioinorganic chemistry. However, problems with toxicity, harsh side effects during administration, and drug resistance have led to increased research to find alternatives to cisplatin and its analogs [1–4].

Carboplatin was the first second-generation analog of cisplatin to be synthesized [5, 6], showing lower toxic side effects [7–9] apparently associated with slow hydrolysis compared to cisplatin, due to possessing a more stable di-carboxylate ligand. Consequently, modification of the labile anionic leaving groups influences the biodistribution and the systemic toxicity. Both cisplatin and carboplatin are effective against the same tumors, due to the fact that once the leaving moieties are released from the platinum center the DNA adducts are undistinguishable [10, 11].

Differences in the nature of the leaving ligands of carboplatin (di-carboxylate) with respect to cisplatin (dichloride) cause major pharmacokinetic differences between them, including stability [5, 6], cytotoxicity [7–9], rates of ligand dissociation by hydroxylation [6, 8, 9], reactivity [12–14], chemotherapeutic dosage [14, 15], plasma proteins and DNA binding kinetics [16], plasma half-life of free (ultrafilterable) platinum, and nephrotoxicity [16, 17].

Like most low molecular weight drugs, cisplatin and carboplatin show a short blood circulation time which reduces tumor uptake and intracellular binding [16, 17]. Therefore, it was envisaged to introduce a limited toxicity and more massive ligand by forming a platinum complex, and thus extending the exposure time of the drug in the body.

These drawbacks prompted us to synthesize a new class of platinum-based anticancer drugs by using a firm molecular platform for the semi-carboplatin cluster. This idea can lead to compounds with enhanced effects and anticancer activities in comparison to the widespread platinum anticancer drug (carboplatin).

Calixarenes have many structural characteristics that are preferable for the design of new drugs. Due to the limited toxicity of calixarenes, they have been broadly used in biology as building blocks or molecular scaffolds, especially as anticancer agents [18–43]. Considering, to date, that calixarenes have showed neither toxicity nor immune responses and that for medical applications the toxicity of molecules is evidently a key factor [18–45], the calixarenes are suitable for use as drug building units.

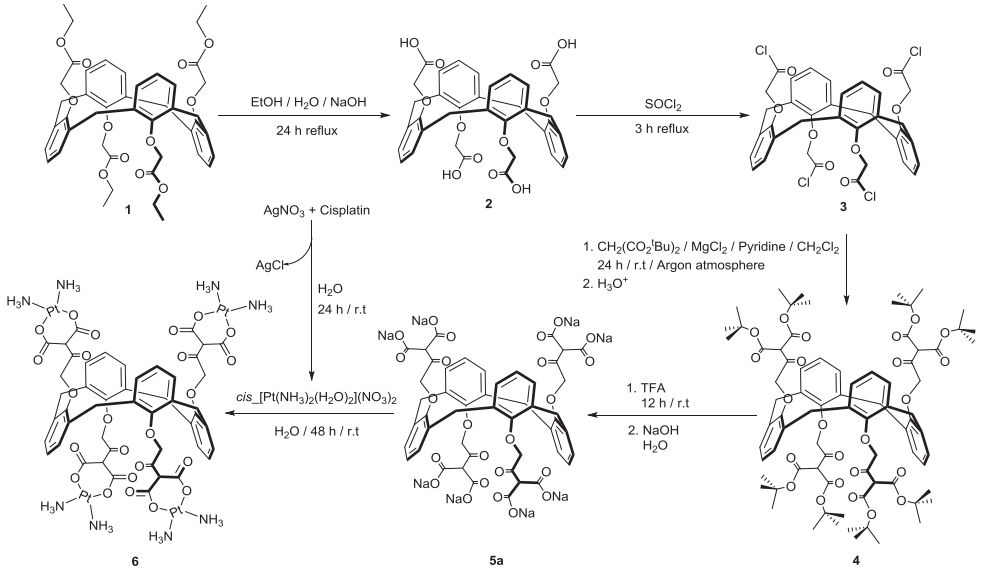
Here, we report the synthesis, characterization, and anticancer activity of a calix[4]arene derivative with four cis-diammineplatinum(II) units attached. The synthetic strategy uses an acetoethoxy spacer on the calix[4]arene to a dicarboxylate.

2. Results and discussion

2.1. *Chemistry*

The synthesis of target compound **6** is shown in scheme 1. For attaching the cis-diammine-platinum(II) units to the calixarene scaffold, an approach was applied in analogy to the synthesis of carboplatin (**B**) by employing a malonic acid motif as the ideal bidentate chelating ligand.

A malonic acid motif could be connected to the calixarene through an acetoethoxy spacer by the following process: the calix[4]arene was refluxed with ethyl 2-bromoacetate and cesium carbonate in dry acetone under an argon atmosphere for 24 h, which gave **1** in a yield of 30% [46]. After that, the tetra-ester was converted to tetra-acid chloride **3** [47] via tetra-acid intermediate **2** [48, 49] by treatment with excess thionyl-chloride. Further reaction with di-tert-butyl malonate, pyridine as base, and $MgCl_2$ in dry dichloromethane under argon for 24 h at ambient temperature afforded **4** in 58% yield. The tert-butyl ester could be smoothly removed with trifluoroacetic acid (TFA) at room temperature for 12 h to give **5** in quantitative yield. The octa-acid **5** was carefully titrated with a 1 N solution of NaOH in a small amount of bi-distilled water till the solid dissolved. Upon addition of methanol to the solution, a white precipitate of the octa-sodium salt **5a** was formed in 72% yield. To enhance ligand bonding, cisplatin (**A**) was converted into the cis-diammine(diaqua)platinum (II) nitrate (activated form) with silver nitrate in bi-distilled water at room temperature after 24 h [50]. The precipitated silver chloride was filtered off and the activated functionalized ligand **5a** in the octa-sodium salt form was added to the solution. After stirring at room temperature for 48 h, the precipitate was formed by adding methanol. It was filtered and washed with cold methanol without further purification to give the target compound **6** in 79% yield.



Scheme 1. Synthetic pathway to calixplatin.

The compounds were characterized by FT-IR, NMR and FAB-MS spectra, and elemental analyses.

The octa-acid **5** was insoluble in DMSO-d_6. So, it was used only for elemental analyses and FAB MS. For complexation, the octa-acid form should be activated and converted to the octa-sodium salt **5a** (water soluble and suitable for NMR spectra) to react with the activated form of cisplatin, cis-diammine(diaqua)platinum(II). NaOH was used to neutralize excess TFA and to deprotonate the octa-acid, which both solubilizes and activates it.

No ^{1}H NMR signal was observed for NH_3 in the target complex. There was a coordination shift of 3.8 ppm for the carboxylate carbons in the ^{13}C NMR spectra of **5a** and **6** (from 182.8 ppm for **5a** to 186.6 ppm for **6**), indicative of complexation of the platinium(II) with **5a** [51]. This coordination shift is comparable to those observed for sodium carboxylates coordinating to form carboplatin derivatives (ranging from 4 to 8.3 ppm) [51].

The binding of the activated compound **5a** to cis-diammine(diaqua)platinum(II) nitrate was confirmed by the shift of $\nu_{C=O}$ (1627 and 1725 cm^{-1} for the free acid form **5**) of the octa-sodium salt form **5a** to lower frequencies for **6** (1604 cm^{-1}), which is comparable to a single band at 1612 cm^{-1} for C=O stretch of carboplatin [52] and the presence of ν_{N-H} (3230–3580 cm^{-1}), as well as an extremely weak band at about 526 cm^{-1} for the symmetric Pt–N stretching vibrations (comparable to 545 cm^{-1} for carboplatin) and the strong band at 464 cm^{-1} (475 cm^{-1} for carboplatin) for the symmetric Pt–O stretch in the FT-IR spectrum of the complex [52].

As expected, the ^{195}Pt NMR spectrum of the complex showed only one signal at -1987 ppm. Compared to cisplatin (−2106 ppm) [53], the coordination of oxygens in the complex (instead of chlorides in cisplatin) resulted in a downfield shift of the signal in the ^{195}Pt NMR spectrum [54]. Most carboplatin derivatives have one ^{195}Pt signal ranging from −1850 to −2000 ppm [54–56].

Poor aqueous solubility is a severe problem for some platinum anticancer drugs in clinical use, such as cisplatin. Hence, the aqueous solubility of calixplatin in comparison to carboplatin was tested at 25 °C. For determining the water solubility of the complexes, they were slowly added to a specified volume of water and addition was continued until saturation was reached. Compared with carboplatin (43 mM L^{-1}, 25 °C), the water solubility of the synthesized complex was improved to 117 mM L^{-1}.

2.2. *In vitro cytotoxic activity*

To determine the half maximal inhibitory growth concentration (IC_{50}) values and the *in vitro* cytotoxicity of calixplatin in comparison to carboplatin as a reference drug, they were evaluated against three different human cancer cell lines. Human carcinoma cell lines were maintained in an RPMI-1640 medium supplemented with 10% fetal bovine serum (FBS), 4 mM glutamine, 100 IU/mL of benzylpenicillin, and 100 µg mL^{-1} of streptomycin. Cells were maintained at 37 °C in a humidified atmosphere of 5% CO_2 in air. The calixplatin and carboplatin were individually pre-dissolved in water and DMSO as stock solutions, respectively. Cells were exposed to a range of 0.1–20 µM of platinum complexes as working solutions. To avoid DMSO toxicity, the concentration of DMSO was less than 0.1% (*v*/*v*) in all experiments. The colorimetric assay of 3-(4,5-dimethylthiazol-2-yl)2,5-diphenyltetrazolium bromide (MTT) was used to establish cell viability by measurement of the absorbance of the resulting dye at 450 nm. The IC_{50} values were calculated from the curves (dose-response curves) constructed by plotting cell survival (%) *versus* complex

Table 1. *In vitro* cytotoxic activity IC_{50} (μM) of calixplatin in comparison to carboplatin against different tumor cell lines (72 h incubation).

Compd.	HepG2	MCF7	A549
Calixplatin	5.7	4.9	2.6
Carboplatin	10.2	15.7	12.8
PtR1* [57]	>100	11.6	26.8
PtR2* [57]	>100	9.5	7.9
PtR3* [57]	>100	22.6	30.1

*Notes: Novel platinum complexes with a new chiral (1*R*,2*R*)-N^1-(pyridine-2-ylmethyl) cyclohexane-1,2-diamine ligand (*R*) and three different leaving groups: $CH_3CO_2^-$ (1), $ClCH_2CO_2^-$ (2), and $CF_3SO_3^-$ (3).

concentration (μM); therefore, the IC_{50} will be equal to the concentration at which cell survival equals 50%.

The results of the cytotoxic activity (IC_{50} (μM)) are summarized in table 1. Calixplatin is more cytotoxic against the different cancer cell lines with lower IC_{50} values than carboplatin, especially for A549 ($IC_{50} = 2.6$ μM).

2.3. *Discussion*

The comparison between the properties of the leaving groups for cisplatin and carboplatin can lead us to find reasons for the enhanced anticancer activity of calixplatin with respect to that of carboplatin as a reference drug.

Replacement of the chloride groups in the cisplatin molecule by the cyclobutanedicarboxylate significantly diminished the nephrotoxic effects of the formed carboplatin, without affecting its antitumor potency [16, 17].

In addition, due to the differences in the nature of the metal–ligand bonds and the chelate effect of the leaving groups, the cyclobutanedicarboxylate bidentate ligand of carboplatin is a much slower leaving group than chloride of cisplatin; hence, cisplatin aquates with a rate constant of 1000-fold more than carboplatin, explaining the higher cytostatic activity of cisplatin [5, 6, 8, 9].

The strategy to reduce toxicity involved increasing the solubility in water and stability of the complexes [4]. Considering that the leaving group in the calixplatin is a bidentate ligand to each of the four platinum(II) sites (functionalized calix[4]arene) that possess higher water solubility with respect to carboplatin, therefore, the property of the new ligand provides a rationale for the reduced toxicity and enhanced anticancer activity of calixplatin as an anticancer agent.

In addition, compared with other synthetic platinum-based anticancer complexes, calixplatin had higher cytostatic activity than PtR1, PtR2, and PtR3 (platinum complexes with a new chiral (1*R*,2*R*)-N^1-(pyridine-2-ylmethyl) cyclohexane-1,2-diamine ligand (R) and three different leaving groups: $CH_3CO_2^-$ (1), $ClCH_2CO_2^-$ (2), and $CF_3SO_3^-$ (3)) against the same human cell lines (table 1) [57–60].

3. Conclusion

In conclusion, the present work describes the first example of a water-soluble platinum–calixarene complex with effective anticancer activity. This derivative could be considered as a novel anticancer agent with enhanced anticancer activity in comparison to carboplatin and

other platinum complexes PtR1, PtR2, and PtR3, especially against the human non-small-cell lung cancer cell line. The enhanced anticancer activity of calixplatin as compared to carboplatin can be attributed to the presence of four platinum units, synergistic effect, the nature of the metal–ligand bonds, and the chelate effect in the complex, in addition to greater water solubility in comparison to carboplatin. The enhanced solubility of the platinum compound in aqueous media probably leads to a reduced cell membrane permeability of the prodrug.

4. Experimental

4.1. *General*

The melting points of all compounds were recorded on a Philip Harris C4954718 apparatus without calibration. IR spectra were determined on a Thermo Nicolet 610 Nexus FT-IR spectrometer with KBr disks. Electronic spectra were scanned on a Shimadzu UV-2401PC. ^{1}H NMR (400 MHz), ^{13}C NMR (100 MHz), and ^{195}Pt NMR (86 MHz) measurements were recorded on a Bruker AM-400 spectrometer. Elemental analyses were obtained on a Perkin–Elmer 240c analyzer. Mass spectra were recorded on a JEOL-JMS 600 (FAB MS) instrument. Thin-layer chromatography analyses were carried out on silica gel plates. All chemicals were purchased from Merck and Aldrich (Tehran, Iran) and used as received. Cell lines were purchased from the American Tissue Culture Collection (ATCC).

4.1.1. 25,26,27,28-Tetrakis(di-tert-butylmalonyl carbonylmethoxy)calix[4]arene (4). $MgCl_2$ (95 mg, 10 mM) was added to dry CH_2Cl_2 (25 mL) under a slight flow of argon. To the resulting heterogeneous mixture, di-*tert*-butyl malonate (4.4 mL, 20 mM) was added. The reaction flask was immersed in an ice bath and pyridine (3 mL) was added via the septum inlet. After the solution was stirred for 15 min at 0 °C, **3** (730 mg, 1 mM) was added. The resulting mixture was stirred 1 h at 0 °C and 24 h at room temperature. After being cooled to 0 °C, the reaction mixture was quenched with 5 N HCl (15 mL), washed with water, dried, and recrystallized from methanol to obtain **4**. Yield (840 mg, 58%), m.p.: 265−267 °C. ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.09 (d, J = 7.3 Hz, 8H, Ar–H_m), 6.70 (t, J = 7.3 Hz, 4H, Ar–H_p), 4.98 (s, 4H, C−H), 4.12 (s, 8H, $ArCH_2Ar$), 3.69 (s, 8H, $ArO-CH_2$), 1.34 (s, 72H, CH_3); ^{13}C NMR (100 MHz, $CDCl_3$) δ 169.5 and 169.1 (C=O), 155.6 (ArC−O), 132.7 ($C_{(o)}$ of Ar), 128.9 ($C_{(m)}$ of Ar), 121.3 ($C_{(p)}$ of Ar), 110.2 ($O\underline{C}^{*}(Me)_3$), 72.1 ($\underline{C}(C=O)_2$), 70.7 ($ArO-CH_2$), 33.9 ($ArCH_2Ar$), 26.9 (CH_3). Anal. Calcd for $C_{80}H_{104}O_{24}$: C, 66.28; H, 7.23. Found: C, 66.42; H, 6.98. FAB^{+} MS *m/z*: 1448.73 (M^{+}).

4.1.2. 25,26,27,28-Tetrakis(di-malonyl carbonylmethoxy)calix[4]arene octasodium salt (5a). Compound **4** (500 mg, 0.35 mM) was suspended in TFA (10 mL) and stirred at room temperature for 12 h. The solvent was then removed under reduced pressure and the residue was triturated with water and filtered to obtain the octa-acid **5** in quantitative yield. After filtration, **5** was carefully titrated with a 1 N solution of NaOH in a small amount of bi-distilled water till the solid dissolved. Upon addition of MeOH to the solution, a white precipitate formed which was collected and dried at 80 °C for 6 h under vacuum to afford octasodium salt **5a**. Yield (250 mg, 72%), m.p. > 350 °C. ^{1}H NMR (400 MHz, DMSO-d_6) δ 7.17 (d, J = 7 Hz, 8H, Ar–H_m), 6.85 (t, J = 7 Hz, 4H, Ar–H_p), 5.01 (s, 4H, C−H), 4.03

(s, 8H, $ArCH_2Ar$), 3.61 (s, 8H, $ArO-CH_2$); ^{13}C NMR (100 MHz, DMSO-d_6) δ 182.8 (O−C=O), 172.5 (C=O), 158.1 (ArC−O), 132.7 ($C_{(o)}$ of Ar), 129.8 ($C_{(m)}$ of Ar), 122.1 ($C_{(p)}$ of Ar), 75.2 ($\underline{C}(C=O)_2$), 68.9 ($ArO-CH_2$), 33.8 ($ArCH_2Ar$). Anal. Calcd for $C_{48}H_{40}O_{24}$: C, 57.61; H, 4.03. Found: C, 57.97; H, 3.92. FAB^+ MS *m/z*: 1000.15 (M^+).

4.1.3. Calixplatin, platinum-calix[4]rene complex (6). Cisplatin (70 mg, 0.23 mM) and $AgNO_3$ (70 mg, 0.42 mM) were dissolved in bi-distilled water (10 mL) and stirred at room temperature for 24 h. After filtration of the precipitated AgCl, the octa-sodium salt **5a** (50 mg, 50 μM) was added and the mixture was stirred at room temperature for 48 h. Precipitate was formed by adding methanol (20 mL). After filtration of the precipitated compound, it was washed with cooled methanol (3 × 10 mL) to give **6**. Yield (75 mg, 79%), m.p.: 186−187 °C. ^1H NMR (400 MHz, D_2O) δ 7.35 (d, J = 7.5 Hz, 8H, Ar–H_m), 6.88 (t, J = 7.5 Hz, 4H, Ar-H_p), 5.22 (s, 4H, C−H), 4.17 (s, 8H, $ArCH_2Ar$), 3.82 (s, 8H, $ArO-CH_2$); ^{13}C NMR (100 MHz, D_2O) δ 186.6 (O−C=O), 173.2 (C = O), 157.1 (ArC−O), 133.2 ($C_{(o)}$ of Ar), 129.0 ($C_{(m)}$ of Ar), 121.7 ($C_{(p)}$ of Ar), 72.3 ($\underline{C}(C=O)_2$), 70.8 ($ArO-CH_2$), 34.2 ($ArCH_2Ar$); ^{195}Pt NMR (86 MHz, D_2O) δ 1987 ppm. Anal. Calcd for $C_{48}H_{56}N_8O_{24}Pt_4$: C, 30.20; H, 2.96; N, 5.87. Found: C, 30.48; H, 3.08; N, 5.66. FAB^+ MS *m/z*: 1908.24 (M^+).

4.2. *Cell lines*

In the present study, cytotoxicity tests were carried out with calixplatin and carboplatin against three different human carcinoma cell lines consisting of A549 [ATCC CCL-185] (human non-small-cell lung cancer cell line), HepG2 [ATCC HB-8065] (human hepatocellular carcinoma cell), and MCF7 [ATCC HTB-22] (human breast cancer cell line).

4.3. *Cell proliferation assay*

Potential cytotoxicity was evaluated against an *in vitro* panel of three different human cancer cell lines. Compound **6** was pre-dissolved in water and carboplatin was pre-dissolved in DMSO (5 mM for avoiding DMSO toxicity) and diluted with cell culture medium to six required concentrations (0.1, 0.2, 1, 2, 10, and 20 μM). All cells were cultured in RPMI-1640 supplemented with 10% FBS, 4 mM glutamine, 100 IU/mL benzylpenicillin, and 100 μg mL^{-1} streptomycin at 37 °C in a humidified atmosphere of 5% CO_2 for 24 h. The cells were seeded at a density of 5×10^4 cells per well in 96-well microplates. After 48 h, the cells were treated with a serial concentration of the test compound. The cells were exposed to drugs in microplates, which were incubated under tissue culture conditions for 72 h. Cell growth was assayed using the colorimetric MTT assay by measurement of the optical density at a wavelength of 450 nm by a microplate reader. Values are mean for data from at least three independent experiments with quadruplicate readings in each experiment.

References

[1] D. Gibson. *Dalton Trans.*, 10681 (2009).
[2] M. Galanski, B.K. Keppler. *Anti-Cancer Agent Med. Chem.*, **7**, 55 (2007).
[3] N.J. Wheate, S. Walker, G.E. Craig, R. Oun. *Dalton Trans.*, 8113 (2010).
[4] A.S. Abu-Surrah, M. Kettunen. *Curr. Med. Chem.*, **13**, 1337 (2006).
[5] B.D. Evans, K.S. Raju, A.H. Calvert, S.J. Harland, E. Wiltshaw. *Cancer Treat. Rep.*, **67**, 997 (1983).

[6] R.B. Weiss, M.C. Christian. *Drugs*, **46**, 360 (1993).
[7] L.J. Ettinger, M.D. Krailo, P.S. Gaynon, G.D. Hammond. *Cancer*, **72**, 917 (1993).
[8] B.J. Foster, K. Clagett-Carr, B. Leyland Jones, D. Hoth. *Cancer Treat. Rev.*, **12**, 43 (1985).
[9] U. Frey, J.D. Ranford, P.J. Sadler. *Inorg. Chem.*, **32**, 1333 (1993).
[10] D. Lebwohl, R. Canetta. *Eur. J. Cancer*, **34**, 1522 (1998).
[11] O. Rixe, W. Ortuzar, M. Alvarez. *Biochem. Pharmacol.*, **52**, 1855 (1996).
[12] J. Reedijk. *J. Chem. Commun.*, **7**, 801 (1996).
[13] L.R. Kelland. *Crit. Rev. Oncol. Hematol.*, **15**, 191 (1993).
[14] P.J. Kuppen, H. Schuitemaker, L.J. Van't Veer, E.A. De Bruijn, A.T. Van Oosterom, P.I. Schrier. *Cancer Res.*, **48**, 3355 (1988).
[15] P. Bedford, S.A. Shellard, M.C. Walker, R.D.H. Whelan, J.R.W. Masters, B.T. Hill. *Int. J. Cancer*, **40**, 681 (1987).
[16] Z.H. Siddik, D.R. Newell, F.E. Boxall, K.R. Harrap. *Biochem. Pharmacol.*, **36**, 1925 (1987).
[17] J. Lokich, N. Anderson. *Annals Oncol.*, **9**, 13 (1998).
[18] M.A. Blaskovich, Q. Lin, F.L. Delarue, J. Sun, H.S. Park, D. Coppola, A.D. Hamilton, S.M. Sebti. *Nat. Biotechnol.*, **18**, 1065 (2000).
[19] H. Zhou, D. Wang, L. Baldini, E. Ennis, R. Jain, A. Carie, S.M. Sebti, A.D. Hamilton. *Org. Biomol. Chem.*, **4**, 2376 (2006).
[20] J. Cai, B.A. Rosenzweig, A.D. Hamilton. *Chem. Eur. J.*, **15**, 328 (2009).
[21] L.K. Tsou, G.E. Dutschman, E.A. Gullen, M. Telpoukhovskaia, Y.-C. Cheng, A.D. Hamilton. *Bioorg. Med. Chem. Lett.*, **20**, 2137 (2010).
[22] S.J. Kim, B.H. Kim. *Tetrahedron Lett.*, **43**, 6367 (2002).
[23] C. Geraci, G.M.L. Consoli, E. Galante, E. Bousquet, M. Pappalardo, A. Spadaro. *Bioconjugate Chem.*, **19**, 751 (2008).
[24] G.M.L. Consoli, G. Granata, E. Galante, I. Di Silvestro, L. Salafia, C. Geraci. *Tetrahedron*, **63**, 10758 (2007).
[25] K. Bezouska, R. Snajdrova, K. Krenek, M. Vancurova, A. Kadek, D. Adamek, P. Lhotak, D. Kavan, K. Hofbauerova, P. Man, P. Bojarova, V. Kren. *Bioorg. Med. Chem.*, **18**, 1434 (2010).
[26] X. Chen, R.P.M. Dings, I. Nesmelova, S. Debbert, J.R. Haseman, J. Maxwell, T.A. Hoye, K.H. Mayo. *J. Med. Chem.*, **49**, 7754 (2006).
[27] F. Sansone, M. Dudic, G. Donofrio, C. Rivetti, L. Baldini, A. Casnati, S. Cellai, R. Ungaro. *J. Am. Chem. Soc.*, **128**, 14528 (2006).
[28] M. Dudic, A. Colombo, F. Sansone, A. Casnati, G. Donofrio, R. Ungaro. *Tetrahedron*, **60**, 11613 (2004).
[29] V. Bagnacani, F. Sansone, G. Donofrio, L. Baldini, A. Casnati, R. Ungaro. *Org. Lett.*, **10**, 3953 (2008).
[30] S. Andre, F. Sansone, H. Kaltner, A. Casnati, J. Kopitz, H.-J. Gabius, R. Ungaro. *ChemBioChem*, **9**, 1649 (2008).
[31] M. Mourer, H.M. Dibama, S. Fontanay, M. Grare, R.E. Duval, C. Finance, J.-B. Regnouf-De-Vains. *Bioorg. Med. Chem.*, **17**, 5496 (2009).
[32] M. Mourer, N. Psychogios, G. Laumond, A.M. Aubertin, J.-B. Regnouf-De-Vains. *Bioorg. Med. Chem.*, **18**, 36 (2010).
[33] H.M. Dibama, I. Clarot, S. Fontanay, A.B. Salem, M. Mourer, C. Finance, R.E. Duval, J.-B. Regnouf-De-Vains. *Bioorg. Med. Chem. Lett.*, **19**, 2679 (2009).
[34] T. Mecca, F. Cunsolo. *Tetrahedron*, **63**, 10764 (2007).
[35] R.V. Rodik, A.S. Klymchenko, N. Jain, S.I. Miroshnichenko, L. Richert, V.I. Kalchenko, Y. Mely. *Chem. Eur. J.*, **17**, 5526 (2011).
[36] V. Paquet, A. Zumbuehl, E.M. Carreira. *Bioconjugate Chem.*, **17**, 1460 (2006).
[37] A. Marra, L. Moni, D. Pazzi, A. Corallini, D. Bridi, A. Dondoni. *Org. Biomol. Chem.*, **6**, 1396 (2008).
[38] L. Frish, F. Sansone, A. Casnati, R. Ungaro, Y. Cohen. *J. Org. Chem.*, **65**, 5026 (2000).
[39] D. Arosio, M. Fontanella, L. Baldini, L. Mauri, A. Bernardi, A. Casnati, F. Sansone, R. Ungaro. *J. Am. Chem. Soc.*, **127**, 3660 (2005).
[40] G.M.L. Consoli, F. Cunsolo, C. Geraci, V. Sgarlata. *Org. Lett.*, **6**, 4163 (2004).
[41] S. Gordo, V. Martos, E. Santos, M. Menendez, C. Bo, E. Giralt, J. De Mendoza. *Proc. Natl. Acad. Sci. USA*, **105**, 16426 (2008).
[42] S. Cecioni, R. Lalor, B. Blanchard, J.-P. Praly, A. Imberty, S.E. Matthews, S. Vidal. *Chem. Eur. J.*, **15**, 13232 (2009).
[43] S. Safi, Z. Asfari, L. Ehret-Sabatier, M. Leroy, A. Hagege. *Bioconjugate Chem.*, **17**, 1346 (2006).
[44] S. Shinkai, K. Araki, O. Manabe. *J. Chem. Soc., Chem. Commun.*, **3**, 187 (1988).
[45] F. Perret, A.N. Lazar, A.W. Coleman. *Chem. Commun.*, **23**, 2425 (2006).
[46] K. No, H.J. Lee, K.M. Park, S.S. Lee, K.H. Noh, S.K. Kim, J.Y. Lee, J.S. Kim. *J. Heterocycl. Chem.*, **41**, 211 (2004).
[47] F. Arnaud-Neu, G. Barrett, S. Cremin, M. Deasy, G. Ferguson, S.J. Harris, A.J. Lough, L. Guerra, M.A. McKervey, M.J. Schwing-Weill, P. Schwinte. *J. Chem. Soc., Perkin Trans*, **2**, 1119, (1992).
[48] P. Zlatuskova, I. Stibor, M. Kadlecova, P. Lhotak. *Tetrahedron*, **60**, 11383 (2004).
[49] V. Stastny, I. Stibor, H. Dvorakova, P. Lhotak. *Tetrahedron*, **60**, 3383 (2004).

[50] O. Aronov, A.T. Horowitz, A. Gabizon, D. Gibson. *Bioconjugate Chem.*, **14**, 563 (2003).
[51] J. Moker, J. Thiem. *Eur. J. Org. Chem.*, **4842**, (2009).
[52] R. Wysokinski, J. Kuduk-Jaworska, D. Michalska. *J. Mol. Struct. (Theochem)*, **758**, 169 (2006).
[53] C.J. Boreham, J.A. Broomhead, D.P. Fairlie. *Aust. J. Chem.*, **34**, 659 (1981).
[54] M.V. De Almeida, E.T. Cesar, E.C.A. Felicio, A.P.S. Fontes, M. Robert-Gero. *J. Braz. Chem. Soc.*, **11**, 154 (2000).
[55] R.A. Tromp, S.S.G.E. Van Boom, C.M. Timmers, S. Van Zutphen, G.A. Van Der Marel, H.S. Overkleeft, J.H. Van Boom, J. Reedijk. *Bioorg. Med. Chem. Lett.*, **14**, 4273 (2004).
[56] D. Urankar, J. Kosmrlj. *Inorg. Chim. Acta*, **363**, 3817 (2010).
[57] C. Gao, F. Fei, T. Wang, B. Yang, S. Gou, J. Yang, L. Liao. *J. Coord. Chem.*, **66**, 1068 (2013).
[58] L. Li, J. Zhang, L. Ma, Z. Zhang, S. Wang, S. Li, G. Zhou. *J. Coord. Chem.*, **66**, 638 (2013).
[59] J. Zhang, F. Zhang, L. Wang, J. Du, S. Wang, S. Li. *J. Coord. Chem.*, **65**, 2159 (2012).
[60] T.T. Da, L.X. Chien, N.T.T. Chi, L.T.H. Hai, N.H. Dinh. *J. Coord. Chem.*, **65**, 131 (2012).