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Synthesis and Cytotoxicity of Enantiomerically Pure [1,2-Diamino-1-(4-fluorophenyl)-3-methylbutane]platinum(II) Complexes

Anja Dullin,[a] Francois Dufrasne,[b] Michael Gelbcke,[b] and Ronald Gust*[a]

*A series of leaving group derivatives of enantiomerically pure [1,2-diamino-1-(4-fluorophenyl)-3-methylbutane]platinum(II) complexes were synthesized and tested for cytotoxicity. The enantiomeric purity was determined by ^{1}H NMR spectroscopy on the final diamines after derivation with (1*R*)-myrtenal. For coordination to platinum, the diamines were reacted with K_2PtI_4. The treatment of diiodoplatinum(II) complexes (4F-Ph/iProp-PtI_2) with Ag_2SO_4 resulted in the sulfatoplatinum(II) complexes (4F-Ph/iProp-$PtSO_4$), which can be easily transformed to dichloroplatinum(II) complexes (4F-Ph/iProp-$PtCl_2$) with 2 N HCl. The importance of the leaving groups and the configuration at the diamine ligand on the antiproliferative effects was evaluated on the hormone-dependent MCF-7 and the hormone-independent MDA-MB 231 breast cancer cell lines as well as the LNCaP/FGC prostate cancer cell line. (*R,R*)-4F-Ph/iProp-$PtCl_2$ was identified as the most active platinum(II) complex. The 3-methyl group increased antiproliferative effects relative to the [1,2-diamino-1-(4-fluorophenyl)butane]platinum(II) complexes described in an earlier study.*

Introduction

Malignant disease is a major cause of mortality all over the world. The era of chemotherapy commenced in the late 1940s and 1950s with the clinical introduction of the classical alkylating agents such as cyclophosphamide, nitrogen mustard, and melphalan. The discovery of the antitumor effects of cisplatin (*cis*-diamminedichloroplatinum(II)) in 1969 by Rosenberg et al.[1] represents a milestone in cancer treatment. Although cisplatin is routinely used nowadays in the treatment of many tumor diseases such as testicular and ovarian cancer, toxic side effects such as nephrotoxicity and myelosuppression[2] were not tolerable and inspired an intensive search for more-tolerable platinum-based drugs. The second-generation platinum drug carboplatin (*cis*-diamminecyclobutane-1,1′-dicarboxylatoplatinum(II)) went into clinical use in the early 1980s. It possessed decreased side effects and increased water solubility. However, the higher doses needed for cisplatin-like antitumor activity diminished the advantages obtained by the use of the cyclobutane-1,1-dicarboxylate leaving group. Randomized trials of cisplatin versus carboplatin in ovarian cancer demonstrated similar responses and recurrence of tumor growth.[3,4] To overcome this cross-resistance, the so-called third generation of platinum complexes was synthesized. The most promising candidate is oxaliplatin, bearing a 1,2-diaminocyclohexane (DACH) ligand and oxalate as leaving group.[5,6] Today it is used for the treatment of colorectal cancer, for which both cisplatin and carboplatin have been shown to be clinically inactive.

The mode of action of oxaliplatin differs from that of other platinum complexes. It forms fewer DNA adducts than cisplatin, but the DACH carrier ligand may induce DNA lesions which are poorly recognized by DNA repair pathways. This effect depends on the configuration of the ethylenediamine chelate. The *R,R* isomer is more effective than the *S,S* and *R,S*/*S,R* isomers.

This finding induced us to study stereoisomeric antiproliferative effects in the class of [1,2-diamino-1,2-bis(4-fluorophenyl)-ethane]dichloroplatinum(II) (4F-Ph-$PtCl_2$) complexes.[7,8] However, the attempt failed to increase the antiproliferative effects by separation into enantiomerically pure compounds.[9] In an earlier study, we demonstrated the increase of in vitro cytotoxicity by the exchange of one 4-fluorophenyl residue with a methyl (4F-Ph/Me-$PtCl_2$), ethyl (4F-Ph/Et-$PtCl_2$) or propyl group (4F-Ph/Prop-$PtCl_2$).[10–12] Interestingly, the C2-methyl substituent led to different cytotoxic effects of the *threo* isomers: $S,S > R,R > S,R = R,S$.[10] Elongation of the side chain in the *erythro* series with one or two methylene groups also induced this effect: $R,R > S,S > R,S > S,R$.[11,12]

Our aim now is to optimize cytotoxic potency with the use of branched C2-alkyl chains. Herein, we report the synthesis of enantiomerically pure [1,2-diamino-1-(4-fluorophenyl)-3-methylbutane]platinum(II) complexes and their antiproliferative effects on the MCF-7 and MDA-MB 231 breast cancer cell lines as well as on the LNCaP/FGC prostate cancer cell line.

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[a] A. Dullin, Prof. Dr. R. Gust
Institute of Pharmacy, Free University of Berlin
Königin-Luise-Str. 2+4, 14195 Berlin (Germany)
Fax: (+30) 838 56906
E-mail: rgust@zedat.fu-berlin.de

[b] Dr. F. Dufrasne, Dr. M. Gelbcke
Laboratoire de Chimie Pharmaceutique Organique, Institut de Pharmacie
Université Libre de Bruxelles, Campus Plaine 205-5, 1050 Bruxelles, Belgium

Results and Discussion

Synthesis

The *erythro*-configured compounds were obtained using a previously published, modified method reported by O'Brien and Poumellec (Scheme 1).[13]

The synthesis of (1*S*,2*R*)-1,2-diamino-1-(4-fluorophenyl)-3-methylbutane (1*S*,2*R*)-**6** started from (2*R*)-2-amino-3-methylbutanol (2*R*)-**1**, which was first N-protected by two benzyl groups and oxidized to the aldehyde (2*R*)-**3**. The reaction with 4-fluorophenylmagnesium bromide resulted in a mixture of the diastereomeric amino alcohols (1*S*,2*R*)-**4** and (1*R*,2*R*)-**4** in a ratio of 90:10. After separation by column chromatography, the hydroxy group of (1*S*,2*R*)-**4** was activated through mesylate formation. This unstable intermediate could not be isolated because it cyclized readily to an aziridinium ion. Ring opening with N_3^- yielded mainly (1*S*,2*R*)-**5** (93%), which was separated by column chromatography. Reduction and deprotection resulted in the diamine (1*S*,2*R*)-**6** in an overall yield of 33%.

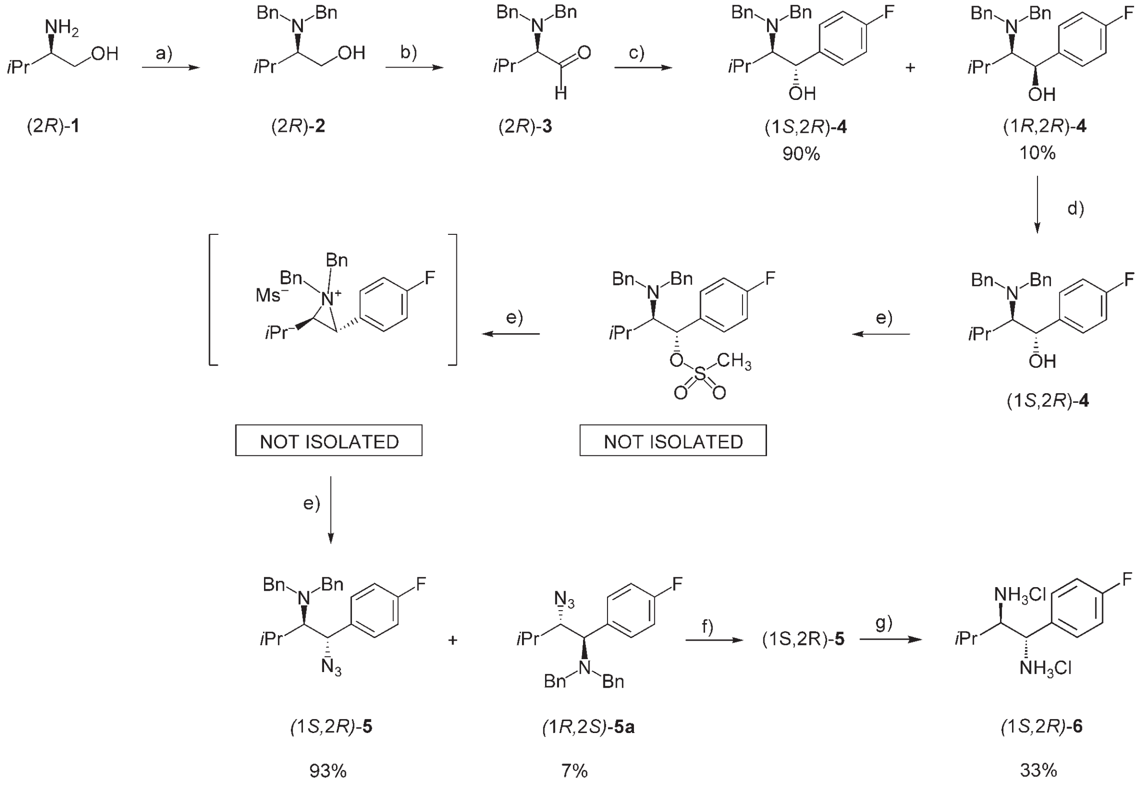
For the preparation of the *threo*-configured diamines, a procedure described by Merino et al. (Scheme 2) was used.[14] (2*R*)-**1** was protected with a *tert*-butoxycarbonyl group to give (2*R*)-**7**, which was oxidized to the aldehyde (2*R*)-**8.** This was allowed to react with *N*-benzylhydroxylamine to form the nitrone (2*R*)-**9**. Addition of 4-fluorophenylmagnesium bromide produced (1*R*,2*R*)-**10** as a diastereomeric mixture with (1*S*,2*R*)-**10** (≈78:22). After separation, (1*R*,2*R*)-**10** was reduced to (1*R*,2*R*)-**11** and deprotected to obtain (1*R*,2*R*)-**12** in an overall yield of 10%.

Using (2*S*)-2-amino-3-methylbutanol (2*S*)-**1**, the diamines (1*R*,2*S*)-**6** and (1*S*,2*S*)-**12** were also made available. In each case, the enantiomeric purity was determined by ^{1}H NMR spectroscopy with (1*R*)-(−)-myrtenal as chiral derivatizing agent.

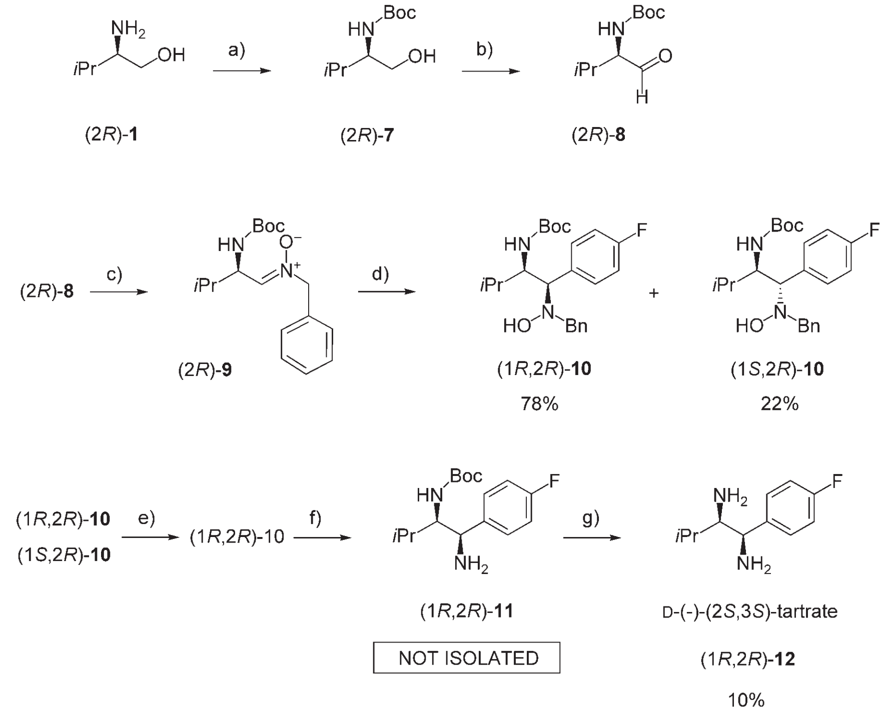
K_2PtI_4, synthesized in situ from K_2PtCl_4 and KI, was used for the coordination of the 1,2-diaminobutanes **6** and **12** (Scheme 3). The reaction with Ag_2SO_4 yielded the respective sulfatoplatinum(II) complexes, which were then transformed to the dichloroplatinum(II) complexes by addition of 2 N HCl. (Scheme 3).

Structural characterization

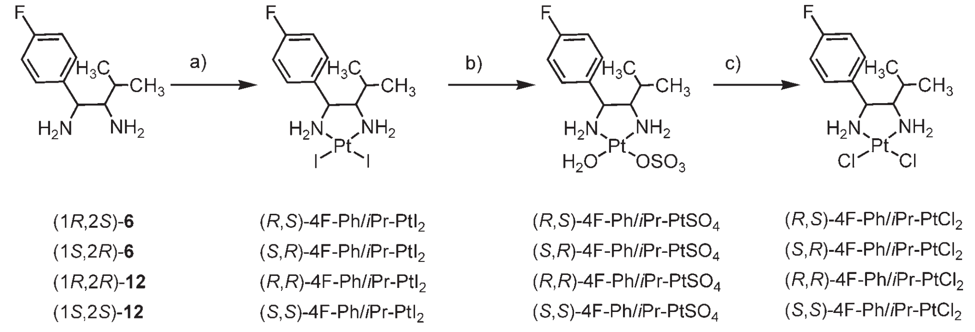
The ^{1}H NMR spectra of the platinum complexes showed characteristic resonances for the amino and methine protons. The protons of both amino groups were diastereomerically split into four signals due to a stable five membered chelate ring at the platinum. If the coordination to platinum was performed in D_2O, a quantitative NH/ND exchange took place and allowed a conformational analysis based on the splitting of the benzylic proton due to the ^3J coupling to the neighboring methine proton and ^{195}Pt. In particular, the platinum satellites resulting



Scheme 1. Synthesis of (1*S*,2*R*)-configured 1,2-diamino-1-(4-fluorophenyl)-3-methylbutane: a) BnBr (2 equiv), KI (0.1 equiv), Na_2CO_3 (2 equiv), THF, reflux; b) DMSO (2 equiv), oxalyl chloride (1.1 equiv), TEA (5 equiv), CH_2Cl_2, −60 °C; c) 4-fluorophenylmagnesium bromide (1.1 equiv), THF, 0 °C; d) column chromatography, petroleum ether$_{(40-60)}$/acetone (9:1); e) 1. MsCl (1.5 equiv), TEA (3 equiv), Et_2O, 0 °C to room temperature; 2. NaN_3 (2 equiv), H_2O; f) column chromatography, petroleum ether$_{(40-60)}$/acetone (98:2); g) 1. $HCOONH_4$ (4 equiv), Pd/C (10%), MeOH, reflux; 2. HCl/Et_2O. Bn = benzyl, Ms = methanesulfonyl.



Scheme 2. Synthesis of (1*R*,2*R*)-configured 1,2-diamino-1-(4-fluorophenyl)-3-methylbutane: a) BOC anhydride (1.1 equiv), TEA (1.1 equiv), CH_2Cl_2, 0 °C to room temperature; b) DMSO (2 equiv), oxalyl chloride (1 equiv), TEA (5 equiv), CH_2Cl_2, −60 °C; c) benzylhydroxylamine (1 equiv), $MgSO_4$ (1 equiv), CH_2Cl_2, room temperature; d) 4F-phenylmagnesium bromide (3 equiv), THF, −50 °C; e) column chromatography, petroleum ether$_{(40-60)}$/acetone (95:5); f) $HCOONH_4$ (4 equiv), Pd/C (10%), MeOH, reflux; g) 1. HCl/MeOH; 2. NaOH; 3. D-(−)-(2*S*,3*S*)-tartaric acid. Bn = benzyl, Boc = *tert*-butoxycarbonyl.



Scheme 3. Synthesis of [1,2-diamino-1-(4-fluorophenyl)-3-methylbutane]platinum(II) complexes: a) K_2PtI_4 (1.1 equiv) at 40 °C in aqueous solution; b) Ag_2SO_4 (0.95 equiv) in aqueous solution with protection from light; c) 2 N HCl.

from the $^3J_{CH-Pt}$ allowed insight into the dynamic effects of the chelate ring.

The benzylic proton of the *threo*-configured compounds showed $^3J_{CH-CH} = 11.8$ Hz and $^3J_{CH-Pt} \approx 0$ Hz. These data were in accordance with a predominant conformation with an equatorially arranged aryl ring.[15] A conversion of the five-membered chelate ring could be excluded. It should be mentioned that the isopropyl residue at the C2 atom was equatorially oriented as well.

The *erythro*-configured compounds can exist in two conformations with either the aryl or the isopropyl residue in an axial position. Then the respective methine resonance possesses platinum satellites at a distance of $^3J = 80$ Hz. An interconversion between both forms is theoretically possible and should increase the $^3J_{CH-CH}$ value and decrease the $^3J_{CH-Pt}$ value.

In the spectra of the *erythro*-configured complexes, the coupling constants of the benzylic proton were $^3J_{CH-CH} = 4.8$ Hz and $^3J_{CH-Pt} = 81$ Hz. This indicated a preference for the conformer with an axially oriented aromatic ring.[15]

Antitumor activity

The antiproliferative activity of the stereoisomeric platinum(II) complexes was determined in tests on the human MCF-7 and MDA-MB 231 breast cancer cell lines, and the LNCaP/FGC prostate cancer cell line. For comparison, cisplatin was also tested. It influenced the proliferation of cell growth in a concentration-dependent manner, and caused 100% inhibition of growth of MCF-7 and MDA-MB 231 cells at the highest concentration (5 μM) (Figure 1). LNCaP/FGC cells were somewhat more resistant against cisplatin (maximal $T/C_{corr} = 20\%$ at 5 μM; Figure 1).

The effects of the [1,2-diamino-1-(4-fluorophenyl)-3-methylbutane]platinum(II) complexes depended on the configuration/conformation of the ethylenediamine ligand and the kind of leaving groups present. The complexes with SO_4^{2-} or Cl^- as leaving groups showed similar cytotoxicity and were by far more active than their diiodoplatinum(II) derivatives. The *threo*-configured complexes were more active than the *erythro*-configured congeners. The separated enantiomers differed in their antiproliferative activity depending on the cell line used.

The (*S*,*S*)- and (*R*,*R*)-4F-Ph/*i*Prop-PtI$_2$ complexes showed similar cytostatic effects on the MCF-7 cell line at a concentration of 5 μM during the whole incubation time of 220 h: $T/C_{corr} = 0\%$ (Figures 2 and 3). At lower concentrations (*R*,*R*)-4F-Ph/*i*Prop-PtI$_2$ was slightly more active than (*S*,*S*)-4F-Ph/*i*Prop-PtI$_2$ (Figure 2). The comparison of the time–activity curves determined with the sulfatoplatinum(II) complexes indicates no significant differences between the enantiomers regarding their cytotoxicity.

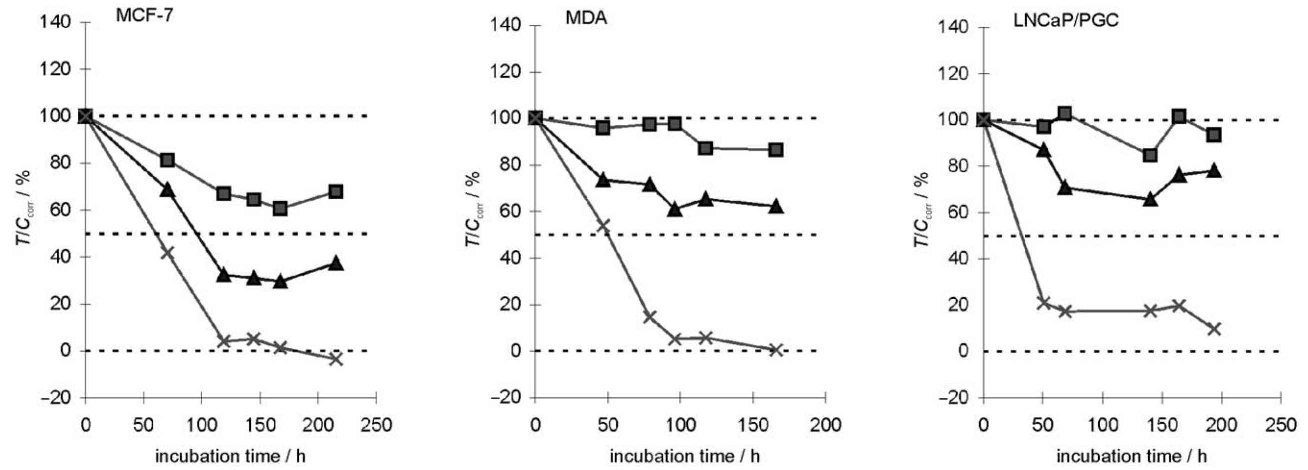


Figure 1. Effect of cisplatin on the MCF-7, MDA-MB 231, and LNCaP/FGC cell lines; ■ = 0.5 μM, ▲ = 1 μM, × = 5 μM.

They caused strong cytocidal effects at a concentration of 5 μM and were cytostatic at a concentration of 1 μM. At 0.5 μM a low recuperation of the cells were observed, somewhat stronger for (*R*,*R*)-4F-Ph/*i*Prop-PtSO$_4$ in comparison with its enantiomer (Figure 2).

The enantiomeric dichloroplatinum(II) complexes showed the most significant differences in cytotoxicity. (*R*,*R*)-4F-Ph/*i*Prop-PtCl$_2$ was the most active compound in this study and caused cytostatic effects at the low concentration of 0.5 μM. Furthermore, it was more active than (*S*,*S*)-4F-Ph/*i*Prop-PtCl$_2$ at each concentration. It should be noted that with the exception of (*S*,*S*)-4F-Ph/*i*Prop-PtI$_2$, all *threo*-configured complexes had similar or greater activity than that of cisplatin.

Among the *erythro* series (Figure 3) (*R*,*S*)- and (*S*,*R*)-4F-Ph/*i*Prop-PtSO$_4$, and (*S*,*R*)-4F-Ph/*i*Prop-PtCl$_2$ showed cisplatin-like effects. Again, the dichloroplatinum(II) complexes possessed significant stereoisomeric differences (cytotoxicity of (*S*,*R*)-4F-Ph/*i*Prop-PtCl$_2$ > (*R*,*S*)-4F-Ph/*i*Prop-PtCl$_2$). Therefore, the tests on the MDA-MB 231 and the LNCaP/FGC cell lines were only performed with these leaving group derivatives.

The growth–activity curves of MDA-MB 231 cells during drug exposure (Figure 4) were characterized by a very fast onset of activity followed by a strong recuperation of the cells. Because exponential cell growth is guaranteed for at least 180 h of incubation, the rise of the growth curve can be explained by the development of drug resistance.[16]

(*S*,*S*)-4F-Ph/*i*Prop-PtCl$_2$ and (*R*,*R*)-4F-Ph/*i*Prop-PtCl$_2$ caused identical cytocidal effects after an incubation period of 48 h at all concentrations tested and a similar recovery of cell growth at the lower concentrations. However at 5 μM,(*R*,*R*)-4F-Ph/*i*Prop-PtCl$_2$ was able to terminate cell division completely for the duration of the test. Interestingly, great differences were observed between the *erythro*-configured compounds. (*R*,*S*)-4F-Ph/*i*Prop-PtCl$_2$ was about fivefold less active than (*S*,*R*)-4F-Ph/*i*Prop-PtCl$_2$, which, in turn, was about fivefold less cytotoxic than (*R*,*R*)-4F-Ph/*i*Prop-PtCl$_2$.

The most promising results were obtained with the LNCaP/FGC prostate cancer cell line (Figure 5). (*S*,*S*)-4F-Ph/*i*Prop-PtCl$_2$ and (*R*,*R*)-4F-Ph/*i*Prop-PtCl$_2$ inhibited cell growth at a very low concentration (0.05 μM), indicating 100-fold higher cytotoxicity than cisplatin. The stereoselectivity was similar to that observed with breast cancer cells: (*R*,*S*)-4F-Ph/*i*Prop-PtCl$_2$ = (*S*,*R*)-4F-Ph/*i*Prop-PtCl$_2$ $\ll$ (*S*,*S*)-4F-Ph/*i*Prop-PtCl$_2$ < (*R*,*R*)-4F-Ph/*i*Prop-PtCl$_2$. Again, (*R*,*R*)-4F-Ph/*i*Prop-PtCl$_2$ was the most active complex.

Discussion

C2-substituted, enantiomerically pure [1,2-diamino-1-(4-fluorophenyl)ethane]platinum(II) complexes are promising new cytotoxic drugs. Their in vitro activity depends on the residue at C2 and the configuration of the 1,2-diaminoethane moiety. These structural features determine the conformational behavior of the five-membered chelate ring and the binding to DNA.

This study was a continuation of our attempts to optimize the antitumor activity of [*S*,*S*/*R*,*R*-1,2-diamino-1,2-bis(4-fluorophenyl)ethane]dichloroplatinum(II) ((*R*,*R*/*S*,*S*)-4F-Ph/4-Ph-PtCl$_2$). (*R*,*R*/*S*,*S*)-4F-Ph/4-Ph-PtCl$_2$ showed high activity in vitro and in vivo against a wide range of murine tumors,[7,8] and was more active than cisplatin and its *R*,*S*/*S*,*R*-configured diastereomeric congener. Separation of the enantiomers did not optimize the antitumor activity of (*R*,*R*/*S*,*S*)-4F-Ph/4-Ph-PtCl$_2$ as the enantiomers show identical pharmacological effects. The separated (*R*,*R*)- and (*S*,*S*)-4F-Ph/4-Ph-PtCl$_2$ possessed identical effects in vitro on the MCF-7 cell line and in vivo on P388 leukemia implanted in mice.[9]

This negative result was not predictable because asymmetric DNA was identified as a target of (*R*,*R*/*S*,*S*)-4F-Ph/4-Ph-PtCl$_2$.[17] The mode of DNA attack should be identical for the enantiomers, resulting in diastereomeric adducts with the chiral DNA that should form intrastrand cross links with different kinetics. However, this last step in the reaction with the target seems to be identical for the enantiomers. We ascribed this phenomenon to the equal substitution of the asymmetric C atoms. Therefore, we tried to optimize the activity of 4F-Ph/4-Ph-PtCl$_2$ complexes by exchanging one aromatic ring with alkyl chains of various lengths.

This structural modification increased the cytotoxicity observed with the human MCF-7 and MDA-MB 231 cell lines (4-fluorophenyl ≤ methyl < ethyl ≈ propyl ≈ butyl). The maximal effect was observed with the [1,2-diamino-1-(4-fluorophenyl)-butane]platinum(II) complex ((*R*,*R*/*S*,*S*)-4F-Ph/Et-$PtCl_2$), the cytotoxicity of which could be optimized by separation of the enantiomers: (*R*,*R*)-4F-Ph/Et-$PtCl_2$ showed a significantly higher activity than (*S*,*S*)-4F-Ph/Et-$PtCl_2$. The same enantioselectivity was observed for the *erythro* compounds: (*R*,*S*)-4F-Ph/Et-$PtCl_2$ was more active than (*S*,*R*)-4F-Ph/Et-$PtCl_2$.[11]

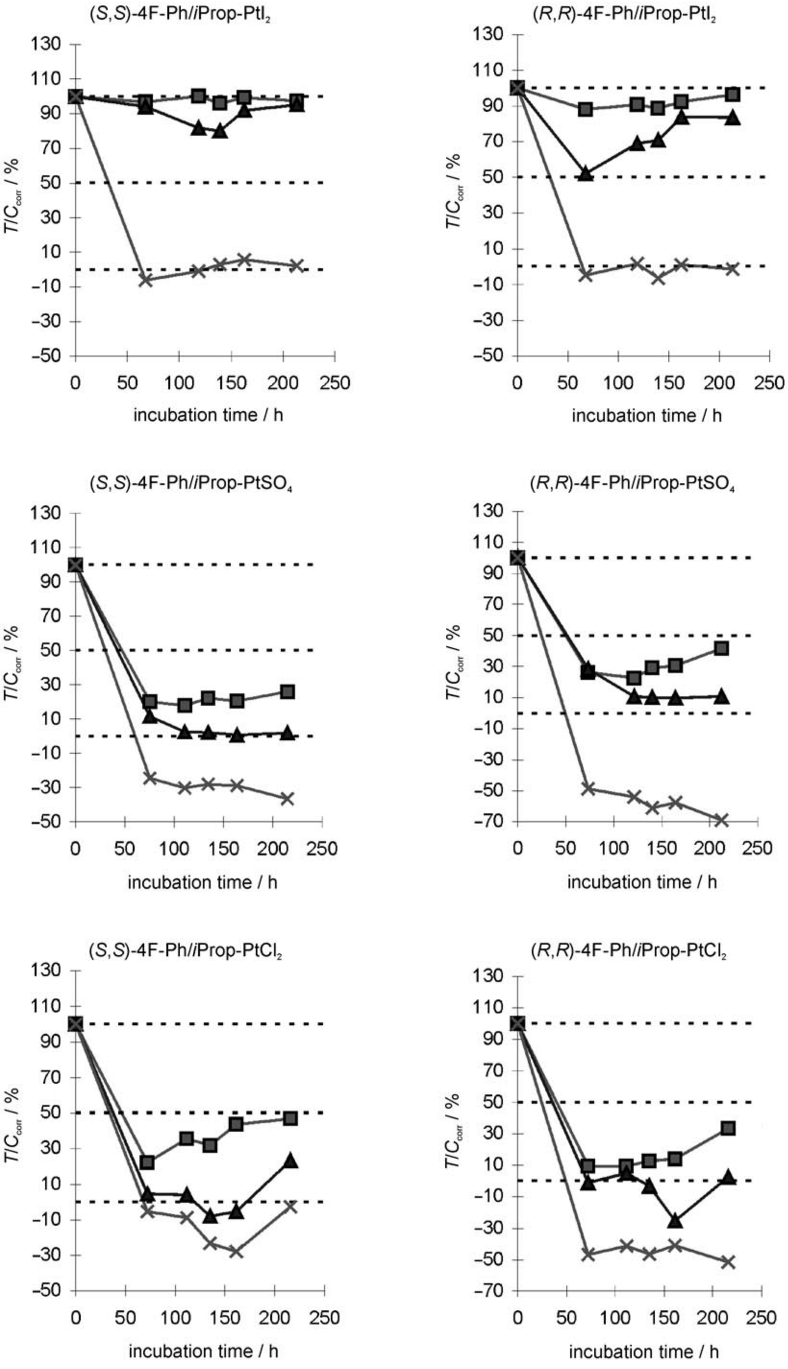


Figure 2. Effect of *threo*-configured [1,2-diamino-1-(4-fluorophenyl)-3-methylbutane]platinum(II) complexes on the MCF-7 breast cancer cell line; ■ = 0.5 μM, ▲ = 1 μM, × = 5 μM.

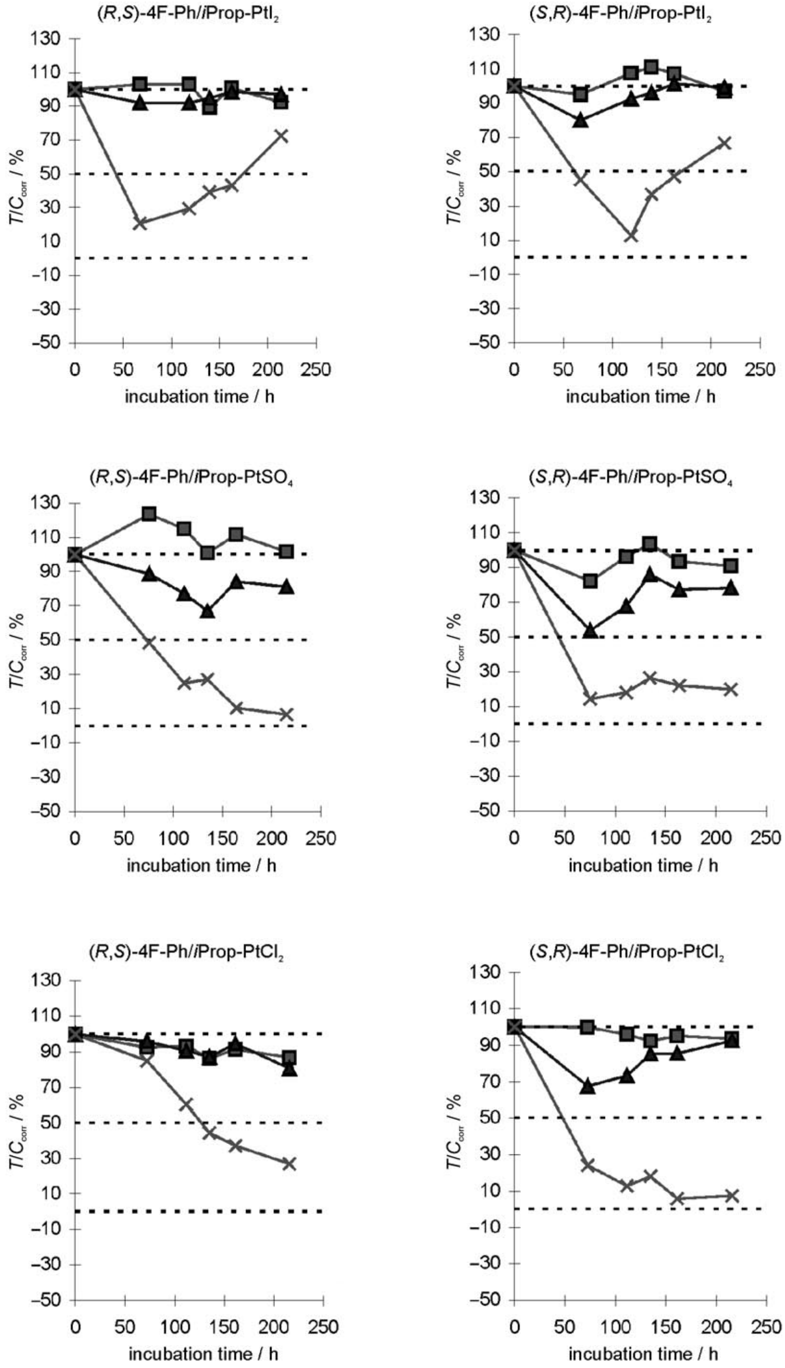
The lack of enhancement of the cytotoxicity by elongation of the ethyl chain prompted us to investigate the influence of a branched isopropyl group at C2 on the antiproliferative effects. This structural modification only marginally affected the activity of the *threo*-configured complexes on the MCF-7 cell line (Figure 2) but was of great significance for their effects on the MDA-MB 231 cell line (Figure 4). In contrast with (*R*,*R*)- and (*S*,*S*)-4F-Ph/Et-$PtCl_2$, strong cytocidal effects were observed for the respective C2-isopropyl derivatives, whereby (*R*,*R*)-4F-Ph/*i*Prop-$PtCl_2$ maintained the cytocidal effects at a concentration of 5 μM for the duration of the experiment.

A more pronounced enantioselectivity was observed in the *erythro* series: (*S*,*R*)-4F-Ph/*i*Prop-$PtCl_2$ was distinctly more active than (*R*,*S*)-4F-Ph/*i*Prop-$PtCl_2$, again to a greater extent on the MDA-MB 231 cell line, on which (*S*,*R*)-4F-Ph/*i*Prop-$PtCl_2$ caused cytocidal effects even at the low concentration of 1 μM. In comparison, the C2-ethyl derivative (*S*,*R*)-4F-Ph/Et-$PtCl_2$ was nearly inactive ($T/C_{corr} \approx 70\%$) at this concentration.

This enhanced activity and stronger enantioselectivity observed in the *erythro* series might be the consequence of the dynamic behavior of the five-membered chelate ring induced by the 3-methyl group at the 1,2-diaminobutane moiety. For (*S*,*R*)-4F-Ph/Et-$PtCl_2$, an interconversion of the chelate ring was verified with a preference for the conformation with the axially oriented aromatic ring. The platinum satellites of $J_{CH-Pt} = 60$ Hz indicated a portion of about 75% at room temperature. In the case of (*S*,*R*)-4F-Ph/*i*Prop-$PtCl_2$, the coupling-constant analyses demonstrated that the isopropyl group forced the aromatic ring in an axial position in a nearly quantitative manner.

It should be mentioned that in the *threo* series only one stable conformation was observed with the alkyl chain and the aromatic ring in an equatorial position. We assume that the conformational behavior of the five-membered ring is highly relevant for DNA binding. The *threo*-configured complexes could easily be bound to the nucleobases because of their relatively flat conformation. The ethyl group in (*S*,*R*)-4F-Ph/Et-$PtCl_2$, and to a much greater extent, the isopropyl group in (*S*,*R*)-4F-Ph/*i*Prop-$PtCl_2$ force the aromatic ring into an axial position. On the one hand, this might hinder DNA binding, but on the other, it is very likely that the DNA lesion is more pronounced after bifunctional binding owing to the predominant axial position of the aromatic ring.

It is well known that increased bulk can alter the interactions of proteins and enzymes with DNA adducts and can influence the nature and lifetime of conformations at the Pt–d(GpG) binding site. In this way, the carrier ligand could affect the anti-


(*R*,*S*)-4F-Ph/*i*Prop-PtI_2

(*S*,*R*)-4F-Ph/*i*Prop-PtI_2

(*R*,*S*)-4F-Ph/*i*Prop-$PtSO_4$

(*S*,*R*)-4F-Ph/*i*Prop-$PtSO_4$

(*R*,*S*)-4F-Ph/*i*Prop-$PtCl_2$

(*S*,*R*)-4F-Ph/*i*Prop-$PtCl_2$

Figure 3. Effect of *erythro*-configured [1,2-diamino-1-(4-fluorophenyl)-3-methylbutane]platinum(II) complexes on the MCF-7 breast cancer cell line; ■ = 0.5 µM, ▲ = 1 µM, × = 5 µM.

cancer activity, the tumor selectivity, and the cross-resistance to cisplatin.[18]

To determine if [1,2-diamino-1-(4-fluorophenyl)-3-methylbutane]platinum(II) complexes are able to overcome cisplatin resistance, we used the LNCaP/FGC prostate cancer cell line as a model. Cisplatin showed only low antiproliferative effects with a $T/C_{corr} \approx 20\,\%$ at the highest concentration used (5 µM). The time–activity curves in Figure 5 clearly demonstrate that these prostate cancer cells showed an increased sensitivity to our novel platinum complexes. To achieve cisplatin-like effects, only 0.05 µM (*S*,*S*)- and (*R*,*R*)-4F-Ph/*i*Prop-$PtCl_2$ were necessary.

The greater potency of the [1,2-diamino-1-(4-fluorophenyl)-3-methylbutane]platinum(II) complexes in LNCaP/FGC cells could be the result of increased cellular uptake, increased DNA adduct levels, or decreased DNA damage tolerance in comparison with cisplatin. Currently, the determination of these pharmacological parameters is under investigation, and the results will be presented in a forthcoming paper.

Experimental Section

^1H and ^{13}C NMR spectra were taken at 293 K on a Bruker Avance 300 MHz spectrometer with TMS as internal standard. IR analysis was performed with a Shimadzu IR-470 spectrophotometer. Melting points (uncorrected) were measured with a Mettler FP1 apparatus. All CC purifications were done using silica gel Kieselgel 100 (Merck). TLC was performed on Kieselgel 60 GF_{254} plates (Merck). Mass spectra were recorded on a Thermo-Fisons VG Auto Spec (70 eV). Specific rotations were measured with a Perkin–Elmer 141 polarimeter.

Experimental procedures were performed with either (2*R*)- or (2*S*)-2-amino-3-methylbutanol ((2*R*)-**1** or (2*S*)-**1**) as starting material. The synthesis is described with (2*R*)-**1** as an example.

(2*R*)-*N*,*N*-dibenzyl-2-amino-3-methylbutanol ((2*R*)-2): Na_2CO_3 (20.6 g, 0.194 mol), KI (1.6 g, 9.7 mmol), and benzyl bromide (33.2 g, 0.194 mol) were added to a solution of (2*R*)-2-amino-3-methylbutanol ((2*R*)-**1**) (10 g, 97 mmol) in THF (200 mL). The suspension was stirred at reflux for 10 h. After cooling and filtration, the solvent was evaporated under reduced pressure. The residue was dissolved in CH_2Cl_2 (200 mL). This solution was washed with brine (50 mL), dried over $MgSO_4$, filtered, and evaporated under reduced pressure. Yield: 27.4 g (100 %) of a yellow oil. ^1H NMR ($CDCl_3$) (base): $\delta = 0.87$ (d, 3 H, $J = 6.6$ Hz, CH_3), 1.12 (d, 3 H, $J = 6.7$ Hz, CH_3), 2.05 (m, 1 H, $(CH_3)_2CH$), 2.52 (m, 1 H, CHN), 2.97 (bs, 1 H, OH), 3.43 and 3.56 (dd and dd, 1 H and 1 H, $J = 4.6$ and 10.7 Hz, CH_2OH), 3.67 and 3.86 (d and d, 2 H and 2 H, $J = 13.2$ Hz, $ArCH_2$), 7.18–7.37 ppm (10 H, Ar). IR (film) (base): $\tilde{\nu} = 3425$ (ν O–H), 2923, 1444, 1095, 1062, 738 (δ Ar), 692 cm^{-1} (δ Ar). $[\alpha]_D^{20}$: (base): (2*R*)-**2**: −23.7 ($c = 0.9$, MeOH).

(2*R*)-*N*,*N*-dibenzyl-2-amino-3-methylbutanal ((2*R*)-3): Oxalyl chloride (13.3 g, 0.105 mol) was dissolved in dry CH_2Cl_2 (100 mL) under N_2 atmosphere and cooled to −60 °C. At this temperature a solution of anhydrous DMSO (14.8 g, 0.19 mol) in dry CH_2Cl_2 (25 mL) was added. After 2 min of stirring, a solution of (2*R*)-*N*,*N*-dibenzyl-2-amino-3-methylbutanol ((2*R*)-**2**) (27 g, 95 mmol) in dry CH_2Cl_2 (50 mL) was added dropwise over 5 min. This mixture was stirred for 15 min, and TEA (48 g, 0.475 mol) was added. The suspension formed was stirred at ∼ −30 °C for 5 min and then allowed to warm to room temperature. Water (200 mL) was added, and the mixture was decanted. The organic layer was washed with brine (50 mL), dried over $MgSO_4$, filtered, and evaporated under reduced pressure. The crude product (brown oil; yield: 23.5 g; 88 %) was not purified before analysis and was used directly for the next

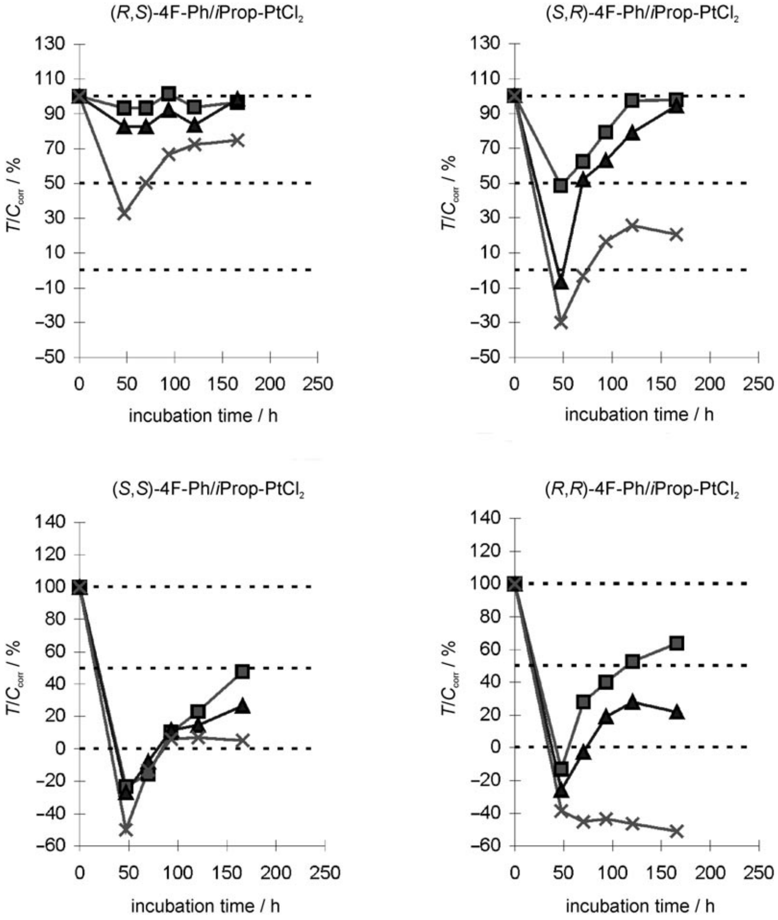


Figure 4. Effect of [1,2-diamino-1-(4-fluorophenyl)-3-methylbutane]dichloroplatinum(II) complexes on the MDA-MB 231 breast cancer cell line; ■ = 0.5 μM, ▲ = 1 μM, × = 5 μM.

step. ^{1}H NMR ($CDCl_3$) (base): $\delta = 0.86$ (d, 3H, $J = 6.6$ Hz, CH_3), 1.07 (d, 3H, $J = 6.7$ Hz, CH_3), 2.28 (m, 1H, $(CH_3)_2CH$), 2.72 (dd, 1H, $J = 3.3$ and 9.9 Hz, CHN), 3.70 and 4.01 (d and d, 2H and 2H, $J = 13.7$ Hz, $ArCH_2$), 7.20–7.38 (10H, Ar), 9.83 ppm (s, 1H, CH=O). IR (film) (base): $\tilde{\nu} = 2805$ (ν C–H), 1717 (ν C=O), 1440, 745 (δ Ar), 692 cm^{-1} (δ Ar).

(1*S*,2*R*)-*N*,*N*-dibenzyl-2-amino-1-(4-fluorophenyl)-3-methylbutanol ((1*S*,2*R*)-4): A 1 M solution of 4-fluorophenylmagnesium bromide in THF (78 mL, 78 mmol) was added dropwise to a solution of (2*R*)-*N*,*N*-dibenzyl-2-amino-3-methylbutanal ((2*S*)-**3**) (20 g, 71 mmol) at 0 °C. The solution was stirred for 2 h at 0 °C and then poured into a saturated NH_4Cl solution (100 mL). The mixture was extracted twice with diethyl ether (100 mL). The organic layers were collected, washed with brine (50 mL), dried over $MgSO_4$, filtered, and evaporated under reduced pressure (viscous yellow oil, 26.7 g). The diastereomeric mixture (90:10) was separated by column chromatography (petroleum ether$_{40-60}$/acetone 95:5). Yield: 18.8 g (70%) of a light yellow viscous oil. ^{1}H NMR ($CDCl_3$) (base): $\delta = 0.96$ (d, 3H, $J = 6.6$ Hz, CH_3), 1.17 (d, 3H, $J = 6.8$ Hz, CH_3), 2.31 (m, 1H, $(CH_3)_2CH$), 2.74 (m, CHN), 3.57 and 3.73 (d and d, 2H and 2H, $J = 13.7$ Hz, $ArCH_2$), 5.04 (d, 1H, $J = 4.0$ Hz, ArCH), 6.94 (m, 2H, ArH3′H5′), 7.14–7.37 ppm (12H, Ar). IR (film) (base): $\tilde{\nu} = 3206$ (ν O–H), 2912 (ν C–H), 1449, 1221 (ν C–F), 843 (δ ArF), 739 (δ Ar), 691 cm^{-1} (δ Ar).

(1*S*,2*R*)-*N*,*N*-dibenzyl-2-amino-1-azido-1-(4-fluorophenyl)-3-methylbutane ((1*S*,2*R*)-5): (1*S*,2*R*)-*N*,*N*-dibenzyl-2-amino-1-(4-fluorophenyl)-3-methylbutanol ((1*S*,2*R*)-**4**) (18 g, 48 mmol) and TEA (14.5 g, 0.143 mol) were dissolved in diethyl ether (200 mL) and cooled with an ice bath. Methanesulfonyl chloride (8.25 g, 72 mmol) was added dropwise. The mixture was stirred for 10 h at room temperature. Water (100 mL) and sodium azide (6.24 g, 96 mmol) were added. After 12 h, the organic layer was taken up, washed with brine (50 mL), dried over $MgSO_4$, and evaporated in vacuo. The mixture of regioisomers ((1*S*,2*R*)-**5** and (1*R*,2*S*)-**5a**) (93:7) was separated by column chromatography (petroleum ether$_{40-60}$/acetone (98:2)). Yield: 10 g (52%) of a viscous yellowish oil. ^{1}H NMR ($CDCl_3$) (base): $\delta = 0.95$ (d, 3H, $J = 6.8$ Hz, CH_3), 1.13 (d, 3H, $J = 6.9$ Hz, CH_3), 2.30 (m, 1H, $(CH_3)_2CH$), 2.75 (dd, 1H, $J = 3.6$ and 7.8 Hz, $CHNBn_2$), 3.51 and 3.58 (d and d, 2H and 2H, $J = 13.5$ Hz, $ArCH_2$), 4.27 (d, 1H, $J = 7.7$ Hz, ArCH), 6.99–7.08 (m, 8H, Ar), 7.02–7.28 ppm (m, 6H, ArH2′H6′). IR (film) (base): $\tilde{\nu} = 2910$ (ν C–H), 2073 (ν N_3), 1585, 1492, 1443, 1219 (ν C–F), 820 (δ ArF), 735 (δ Ar), 698 cm^{-1} (δ Ar).

(1*S*,2*R*)-1,2-diamino-1-(4-fluorophenyl)-3-methylbutane ((1*S*,2*R*)-6): Palladium on charcoal 10% (0.9 g) was added to a solution of (1*S*,2*R*)-*N*,*N*-dibenzyl-2-amino-1-azido-1-(4-fluorophenyl)-3-methylbutane ((1*S*,2*R*)-**5**) (9 g, 22 mmol) in MeOH (75 mL). This suspension was stirred at reflux for 30 min. Then, ammonium formate (2.8 g, 44 mmol) was added, and the mixture was stirred for 10 h. An equal amount of ammonium formate was added, and the reaction was continued for 10 h. The reaction medium was filtered on celite, and the solvent evaporated under reduced pressure. The oily residue was dissolved in CH_2Cl_2 (100 mL). This solution was washed with a solution of sodium carbonate 20% (*w*/*v*) (50 mL) and brine (50 mL). It was further dried over $MgSO_4$, filtered, and evaporated under reduced pressure to give 2.9 g of yellow oil. Upon addition of HCl in diethyl ether, a yellowish solid precipitated. It was filtered and recrystallized from *i*PrOH/MeOH/Et_2O (5:5:20). Yield: 3.84 g (65%) of a colorless solid (hydrochloride); mp: 267 °C with some decomposition. Enantiomeric excess for the two enantiomers: 98% *ee*. ^{1}H NMR ([D_6]DMSO) (hydrochloride): $\delta = 0.96$ (d, 3H, $J = 6.7$ Hz, CH_3), 1.12 (d, 3H, $J = 6.6$ Hz, CH_3), 1.99 (m, 1H, $(CH_3)_2CH$), 3.60 (m, 1H, CHCHN), 4.67 (d, 1H, $J = 5.3$ Hz, ArCH), 7.34 (m, 2H, ArH3′H5′), 7.80 (m, 2H, ArH2′H6′), 8.32 and 9.25 ppm (bs and bs, 3H and 3H, NH_3^+). ^{13}C NMR ([D_6]DMSO) (hydrochloride): $\delta = 14.1$ (CH_3), 19.3 (CH_3), 27.0 ($(CH_3)_2CH$), 54.6 (CHCHN), 57.5 (ArCH), 115.7 (d, $J_{C-F} = 21$ Hz, ArC3′C5′), 130.2 (d, $J_{C-F} = 3$ Hz, ArC1′), 130.5 (d, $J_{C-F} = 8$ Hz, ArC2′C6′), 162.1 ppm (d, $J_{C-F} = 246$ Hz, ArC4′). IR (KBr, 1%) (hydrochloride): $\tilde{\nu} = 2910$ (ν N–H), 2735 (ν C–H), 1502, 1229 (ν C–F), 829 cm^{-1} (δ Ar–F). $[\alpha]_D^{20}$ (hydrochloride) = −23 ($c = 0.3$, MeOH). MS (base): *m*/*z* (%) = 197 (1) [$M+H$], 180 (8), 167 (8), 138 (12), 109 (11), 72 (100), 55 (23).

(2*R*)-*N*-(*tert*-butoxycarbonyl)-2-amino-3-methylbutanol ((2*R*)-7): A solution of (2*R*)-2-amino-3-methylbutanol ((2*R*)-**1**) (10 g, 97 mmol) and TEA (10.8 g, 0.107 mol) in CH_2Cl_2 (50 mL) was cooled to 0 °C. A solution of di-*tert*-butyl dicarbonate (23.35 g, 0.107 mol) in CH_2Cl_2 (10 mL) was added dropwise. The reaction was continued for 12 h at room temperature. The solvents and reagents were evaporated under reduced pressure (60 °C/66 Pa). Yield: 19.7 g (100%) of a yellow oil. ^{1}H NMR ($CDCl_3$): $\delta = 0.93$ (d, 3H, $J = 6.7$ Hz, $(CH_3)_2CH$), 0.95 (d, 3H, $J = 6.7$ Hz, $CH(CH_3)_2$), 1.45 (s, 9H, $C(CH_3)_3$), 1.85 (m, 1H,

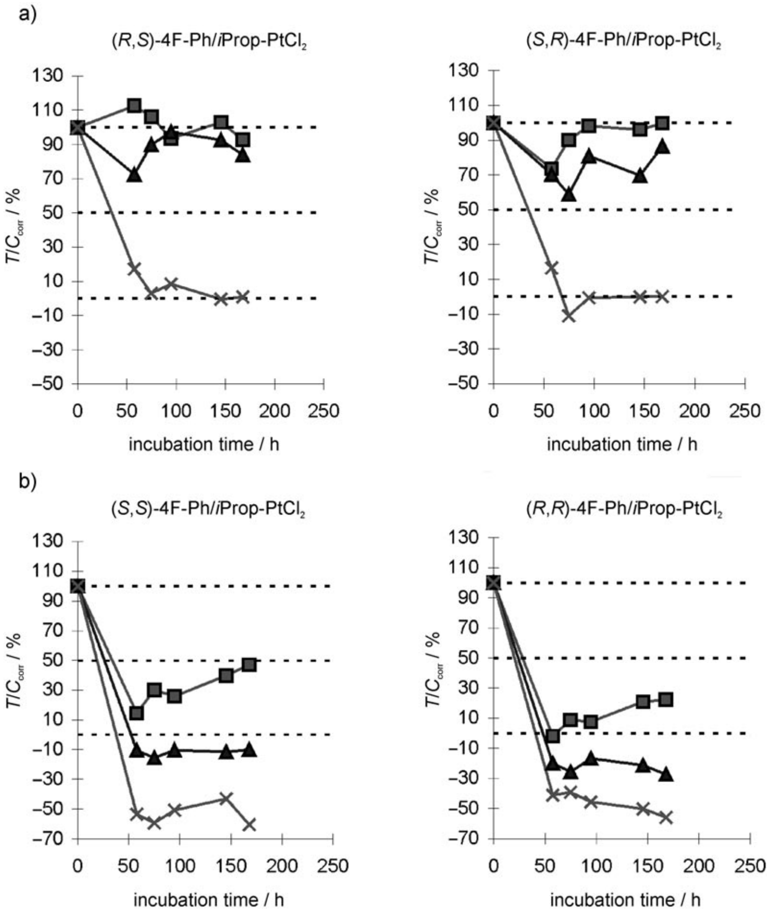


Figure 5. Effect of [1,2-diamino-1-(4-fluorophenyl)-3-methylbutane]dichloroplatinum(II) complexes on the LNCaP/FGC prostate cancer cell line; a) ■ = 0.5 μм, ▲ = 1 μм, × = 5 μм; b) ■ = 0.05 μм, ▲ = 0.1 μм, × = 0.5 μм.

$CH(CH_3)_2$), 3.42 (m, 1H, *C*HN), 3.66 (dd, 2H, *J* = 7.1 Hz and 11 Hz, CH_2OH), 4.85 ppm (d, 1H, *J* = 9.1 Hz, O=CN*H*). ^{13}C NMR ($CDCl_3$): δ = 19.1 (($CH_3)_2CH$), 20.1 (($CH_3)_2CH$), 28.4 (($CH_3)_2CH$), 29.0 ($C(CH_3)_3$), 58.6 (*C*HN), 64.5 (CH_2OH), 80.0 ($C(CH_3)_3$), 157.4 ppm (C=O). IR (film): $\tilde{\nu}$ = 3330 (ν O–H), 2940, 1682 (ν C=O), 1163 cm^{-1}. $[\alpha]_D^{20} = +13.8$ (*c* = 0.3, EtOH).

(2*R*)-(*tert*-butoxycarbonyl)-2-amino-3-methylbutanal ((2*R*)-8): Oxalyl chloride (12.4 g, 98 mmol) in dry CH_2Cl_2 (200 mL) was placed into a three-necked flask under N_2 and cooled to −60 °C. This temperature was maintained during the reaction. Anhydrous DMSO (13.9 g, 0.178 mol) in dry CH_2Cl_2 (50 mL) was added dropwise. After 2 min of stirring, a solution of (2*R*)-*N*-(*tert*-butoxycarbonyl)-2-amino-3-methylbutanol ((2*R*)-**7**) (18 g, 89 mmol) in dry CH_2Cl_2 (50 mL) was added dropwise. After 5 min of stirring, TEA (45 g, 0.445 mol) was added. The suspension was stirred at ~−30 °C for an additional 5 min, and then it was allowed to warm to room temperature. Water (200 mL) was added. After decantation, the organic layer was washed with brine (50 mL), dried over $MgSO_4$, filtered, and evaporated under reduced pressure. The yellow oil (yield: 15.1 g, 85 %) was not purified before analysis and was used directly for the next step. ^1H NMR ($CDCl_3$): δ = 0.95 (d, 3H, *J* = 7.0 Hz, ($CH_3)_2CH$), 1.03 (d, 3H, *J* = 7.0 Hz, ($CH_3)_2CH$), 1.45 (s, 9H, C-($CH_3)_3$), 2.77 (m, 1H, $CH(CH_3)_2$), 4.21 (m, 1H, CHN), 5.13 (bs, 1H, O=CNH), 9.65 (s, 1H, O=CH). ^{13}C NMR ($CDCl_3$): δ = 18.2 (($CH_3)_2CH$), 19.7 (($CH_3)_2CH$), 28.4 (($CH_3)_2CH$), 28.9 ($C(CH_3)_3$), 68.5 (*C*HN), 80.5 ($C(CH_3)_3$), 156.3 (O=*C*NH), 201 (O=*C*H). IR (film): $\tilde{\nu}$ = 3310 (ν N–H), 2941 (ν C–H hydrocarbons), 2705 (ν C–H aldehyde) 1696 large (ν C=O aldehyde and carbamate), 1501, 1158 cm^{-1}.

(2*R*)-[*N*-(*tert*-butoxycarbonyl)-2-amino-3-methylbutylidene]benzylamine *N*-oxide ((2*R*)-9): $MgSO_4$ (9 g, 75 mmol) was added to a solution of (2*R*)-*N*-(*tert*-butoxycarbonyl)-2-amino-3-methylbutanal ((2*R*)-**8**) (15 g, 75 mmol) and benzylhydroxylamine (9.2 g, 75 mmol) in dry CH_2Cl_2 (150 mL). The suspension was stirred for 15 h at room temperature. The solvent was evaporated under reduced pressure after filtration, and the yellow solid obtained was purified by column chromatography (diethyl ether) to afford 14.2 g of a yellow solid (yield: 62 %); mp: 130 °C. ^1H NMR ($CDCl_3$): δ = 0.87 (d, 3H, *J* = 6.7 Hz, ($CH_3)_2CH$), 0.91 (d, 3H, *J* = 6.7 Hz, ($CH_3)_2CH$), 1.41 (s, 9H, $C(CH_3)_3$), 2.28 (m, 1H, ($CH_3)_2CH$), 4.18 (m, 1H, CHNCH=N), 4.86 (s, 2H, $ArCH_2$), 6.00 (bs, 1H, O=CN*H*), 6.78 (bs, 1H, N=CH), 7.37–7.40 ppm (5H, Ar). ^{13}C NMR ($CDCl_3$): δ = 19.7 (($CH_3)_2CH$), 20.2 (($CH_3)_2CH$), 28.4 (($CH_3)_2CH$), 28.9 ($C(CH_3)_3$), 54.8 (*C*HNC=N), 70.4 ($ArCH_2$), 80.1 ($C(CH_3)_3$), 129.6 (ArC2′C6′), 129.7 (ArC3′C4′C5′), 133.3 (ArC1′), 138.23 (C=N), 156.4 ppm (C=O). IR (KBr 1 %): 3320 (ν N–H), 2945 (ν C–H), 1676 (ν C=O, C=N), 1511, 1245, 1165, 696 cm^{-1} (δ Ar).

(1*R*,2*R*)-*N*¹-benzyl-*N*²-(*tert*-butoxycarbonyl)-2-amino-1-(4-fluorophenyl)-1-hydroxyamino-3-methylbutane ((1*R*,2*R*)-10): A solution of (2*R*)-[*N*-(*tert*-butoxycarbonyl)-2-amino-3-methylbutylidene]benzylamine-*N*-oxide ((2*R*)-**9**) (14 g, 46 mmol) in anhydrous THF (100 mL) was cooled to −40 °C and combined dropwise with a 2 м solution of 4-fluorophenylmagnesium bromide in diethyl ether (69 mL, 0.138 mol). Then the reaction medium was stirred at this temperature for 4 h, brought to room temperature, and poured into a saturated solution of NH_4Cl (200 mL). After three extractions with diethyl ether (100 mL), the organic layer was washed with brine (50 mL), dried over $MgSO_4$, filtered, and evaporated under reduced pressure. The brown solid (12 g mixture of diastereomers, 78:22) (1*R*,2*R*)-**10** obtained was separated by column chromatography (petroleum ether$_{(40-60)}$/acetone (95:5)). Yield: 9.7 g (53 %) of a yellow solid; mp: 130.1 °C. ^1H NMR ($CDCl_3$): δ = 0.67 (d, 3H, *J* = 6.8 Hz, ($CH_3)_2CH$), 0.93 (d, 3H, *J* = 6.7 Hz, ($CH_3)_2CH$), 1.49 (m, 1H, ($CH_3)_2CH$), 1.52 (s, 9H, $C(CH_3)_3$), 3.35 (d, 1H, *J* = 10.8 Hz, ArCH), 3.52 and 3.57 (d and d, 1H and 1H, *J* = 14 Hz, $ArCH_2$), 4.11 (ddd, 1H, *J* = 2.7, 10.6, 10.8 Hz, CHC*H*N), 4.55 (d, 1H, *J* = 10.6 Hz, O=CNH), 6.75 (bs, 1H, NOH), 7.06 (m, 2H, ArH3′H5′), 7.18–7.36 ppm (7H, Ar). IR (KBr 1 %): $\tilde{\nu}$ = 3400 (ν N–H O–H), 2945 (ν C–H), 1660 (ν C=O), 1527, 1217 (ν C–F), 1154, 727 cm^{-1} (δ Ar).

(1*R*,2*R*)-*N*²-(*tert*-butoxycarbonyl)-1,2-diamino-1-(4-fluorophenyl)-3-methylbutane ((1*R*,2*R*)-11): (1*R*,2*R*)-*N*¹-benzyl-*N*²-(*tert*-butoxycarbonyl)-2-amino-1-(4-fluorophenyl)-1-hydroxyamino-3-methylbutane ((1*R*,2*R*)-**10**) (9 g, 22 mmol) was dissolved in MeOH (250 mL), and palladium on charcoal 10 % (0.9 g) was added. The suspension was held at reflux for 30 min. Ammonium formate (2.8 g, 45 mmol) was added, and the suspension was kept at reflux for 12 h. Then, an equal amount of ammonium formate was added, and the reaction was continued for 12 h. The cooled reaction mixture was filtered on celite, and the solvent was evaporated under reduced pressure. The residue was extracted with CH_2Cl_2 (100 mL). This solution was washed with brine (50 mL), dried over $MgSO_4$, filtered, and evaporated under reduced pressure. The oily residue (6.5 g) was used directly for the next step.

(1*R*,2*R*)-1,2-diamino-1-(4-fluorophenyl)-3-methylbutane ((1*R*,2*R*)-12): The product of the previous step was brought into a solution of HCl in anhydrous MeOH (10% (*w*/*v*)) (50 mL). This solution was stirred at room temperature for 8 h. Then the solvent was evaporated under reduced pressure. The yellow solid obtained was treated with water (50 mL), NaOH 40% (*w*/*v*) (10 mL), and extracted twice with CH_2Cl_2 (100 mL). The organic solution was washed with brine (50 mL), dried over $MgSO_4$, filtered, and evaporated under reduced pressure. The liquid obtained was dissolved in MeOH (20 mL), and a solution of D-(−)-(2*S*,3*S*)-tartaric acid (3.3 g, 22 mmol) in MeOH (20 mL) was added. This mixture was heated at reflux for 10 min and then it was allowed to cool to room temperature. The white crystals (1.52 g) were collected by filtration, washed twice with MeOH, and dried at 60 °C under 13.3 kPa (yield: 20%). Small fractions of this salt were transformed to base and hydrochloride for analysis; mp: 277.5 °C with some decomposition. Enantiomeric excess for the two enantiomers: 98 %*ee*. ^{1}H NMR ([D_6]DMSO) (hydrochloride): $\delta = 0.75$ (d, 3H, $J = 6.9$ Hz, CH_3), 0.94 (d, 3H, $J = 6.9$ Hz, CH_3), 1.55 (m, 1H, $(CH_3)_2CH$), 3.80 (dd, 1H, $J = 2.5$ and 9.2 Hz, CHCHN), 4.66 (d, 1H, $J = 9.3$ Hz, ArCH), 7.33 (m, 2H, ArH3′H5′), 7.72 (m, 2H, ArH2′H6′), 8.98 ppm (bs, 6H, NH_3^+). ^{13}C NMR ([D_6]DMSO) (hydrochloride): $\delta = 14.1$ (CH_3), 19.3 (CH_3), 27.0 ($(CH_3)_2CH$), 54.6 (CHCHN), 57.6 (ArCH), 115.7 (d, $J_{C-F} = 21$ Hz, ArC3′C5′), 130.2 (d, $J_{C-F} = 3$ Hz, ArC1′), 130.5 (d, $J_{C-F} = 8$ Hz, ArC2′C6′), 162.2 ppm (d, $J_{C-F} = 246$ Hz, ArC4′). IR (KBr, 1%) (hydrochloride): $\tilde{\nu} = 2910$ (ν N–H), 2735 (ν C–H), 1502, 1229 (ν C–F), 829 cm^{-1} (δ Ar–F). $[\alpha]_D^{20}$ (hydrochloride): $= +2.9$ ($c = 0.2$, EtOH). MS (base): m/z (%) = 197 (1) [M+H], 180 (8), 167 (8), 138 (12), 109 (11), 72 (100), 55 (23).

General procedure for the synthesis of [1,2-diamino-1-(4-fluorophenyl-3-methylbutane]diiodoplatinum(II) complexes: A solution of the diamine (0.25 mmol) in 5 mL water was adjusted to pH 6.5–7.5 with 0.1 N NaOH, and combined with K_2PtI_4 (0.25 mmol) dissolved in 5 mL water. The reaction mixture was stirred in the dark for 24 h. Subsequently, it was acidified with 0.1 N HCl, and the yellow precipitate was aspirated and dried over P_2O_5 in vacuo.

(*R*,*R*)-4F-Ph/*i*Prop-PtI_2: 150.00 mg (0.232 mmol, 93%); (*S*,*S*)-4F-Ph/*i*Prop-PtI_2: 147.33 mg (0.228 mmol, 91%): ^{1}H NMR ([D_7]DMF): $\delta = 0.85$ (d, $J = 7.1$ Hz, 3H, CH_3), 0.99 (d, $J = 7.1$ Hz, 3H, CH_3), 1.58 (m, 1H, CH), 3.24 (d, $J = 4.2$ Hz, 1H, CH-alkyl), 3.90 (m, 1H, ArCH), 4.75 (br, 1H, NH), 5.30 (br, 1H, NH), 5.49 (br, 1H, NH), 6.00 (br, 1H, NH), 7.23–7.32 (m, 2H, ArH3′H5′), 7.69–7.78 ppm (m, 2H, ArH2′H6′).(*R*,*S*)-4F-Ph/*i*Prop-PtI_2: 152.00 mg (0.236 mmol, 94%); (*S*,*R*)-4F-Ph/*i*Prop-PtI_2: 149.23 mg (0.231 mmol, 92%): ^{1}H NMR ([D_7]DMF): $\delta = 0.95$–1.04 (m, 6H, CH_3), 1.47 (m, 1H, CH), 3.10–3.13 (m, 1H, CH-alkyl), 4.10 (m, 1H, ArCH), 5.11 (br, 1H, NH), 5.38 (br, 2H, NH), 6.04 (br, 1H, NH), 7.20–7.30 (m, 2H, ArH3′H5′), 8.38–8.49 ppm (m, 2H, ArH2′H6′); elemental analysis for:
$C_{11}H_{17}N_2FPtI_2$ (645.1): calcd: C 20.48, H 2.66, N 4.34;
(*R*,*R*)-4F-Ph/*i*Prop-PtI_2 found: C 20.20, H 2.65, N 4.34,
(*S*,*S*)-4F-Ph/*i*Prop-PtI_2 found: C 20.23, H 2.63, N 4.47,
(*R*,*S*)-4F-Ph/*i*Prop-PtI_2 found: C 20.27, H 2.67, N 4.67,
(*S*,*R*)-4F-Ph/*i*Prop-PtI_2 found: C 20.33, H 2.66, N 4.39.

General procedure for the synthesis of aqua[1,2-diamino-1-(4-fluorophenyl)-3-methylbutane]sulfatoplatinum(II)complexes: The corresponding sulfatoplatinum (II) complex was obtained by addition of Ag_2SO_4 (0.2 mmol) to the aqueous suspension of the respective diiodoplatinum(II) complex (0.2 mmol). The suspension was stirred for 3 days, filtered off, and lyophilized to obtain a white powder.

(*R*,*R*)-4F-Ph/*i*Prop-PtSO_4: 80.00 mg (0.158 mmol, 63%); (*S*,*S*)-4F-Ph/*i*Prop-PtSO_4: 84.21 mg (0.166 mmol, 67%): ^{1}H NMR ([D_6]DMSO): $\delta = 0.70$ (d, $J = 3.5$ Hz, 3H, CH_3), 0.82 (d, $J = 7.2$ Hz, 3H, CH_3), 1.38–1.40 (m, 1H, CH-alkyl), 3.72 (m, 1H, ArCH), 5.82 (br, 1H, NH), 6.19 (br, 1H, NH), 6.30 (br, 1H, NH), 6.44 (br, 1H, NH), 7.22–7.29 (m, 2H, ArH3′H5′), 7.41–7.60 ppm (m, 2H, ArH2′H6′).(*R*,*S*)-4F-Ph/*i*Prop-PtSO_4: 92.31 mg (0.183 mmol, 73%), (*S*,*R*)-4F-Ph/*i*Prop-PtSO_4: 90.00 mg (0.178 mmol, 71%): ^{1}H NMR ([D_6]DMSO): $\delta = 0.87$–0.93 (m, 6H, CH_3), 1.26 (m, 1H, CH), 2.88–2.92 (m, 1H, CH-alkyl), 4.61 (m, 1H, ArCH), 6.28 (br, 1H, NH), 6.43 (br, 2H, NH), 6.68 (br, 1H, NH), 7.32–7.42 (m, 2H, ArH3′H5′), 7.90–7.96 (m, 2H, ArH2′H6′); elemental analysis for:
$C_{11}H_{19}N_2FPtSO_5$ (505.4) calcd: C 26.14, H 3.79, N 5.54;
(*R*,*R*)-4F-Ph/*i*Prop-PtSO_4 found: C 26.08, H 3.53, N 5.46,
(*S*,*S*)-4F-Ph/*i*Prop-PtSO_4 found: C 26.01, H 3.49, N 5.30,
(*R*,*S*)-4F-Ph/*i*Prop-PtSO_4 found: C 25.98, H 3.59, N 5.34,
(*S*,*R*)-4F-Ph/*i*Prop-PtSO_4 found: C 26.08, H 3.60, N 5.44.

General procedure for the synthesis of [1,2-diamino-1-(4-fluorophenyl-3-methylbutane]dichloroplatinum(II) complexes: The respective sulfatoplatinum(II) complex was treated with 2 N HCl and stirred for 6 h. The yellow precipitate was aspirated and dried over P_2O_5.

(*R*,*R*)-4F-Ph/*i*Prop-PtCl_2: 92.30 mg (0.199 mmol, 80%); (*S*,*S*)-4F-Ph/*i*Prop-PtCl_2: 88.76 mmol (0.192 mmol, 77%): ^{1}H NMR ([D_7]DMF): $\delta = 0.88$ (d, $J = 7.0$ Hz, 3H, CH_3), 0.99 (d, $J = 7.1$ Hz, 3H, CH_3), 1.50 (m, 1H, CH), 3.29–3.32 (m, 1H, CH-alkyl), 3.92 (m, 1H, ArCH), 4.98 (br, 1H, NH), 5.49 (br, 1H, NH), 5.63 (br, 1H, NH), 6.10 (br, 1H, NH), 7.21–7.32 (m, 2H, ArH3′H5′), 7.70–7.81 ppm (m, 2H, ArH2′H6′).(*R*,*S*)-4F-Ph/*i*Prop-PtCl_2: 79.30 mg (0.171 mmol, 69%); (*S*,*R*)-4F-Ph/*i*Prop-PtCl_2: 82.45 mg (0.178 mmol, 71%): ^{1}H NMR ([D7]DMF): $\delta = 1.00$–1.03 (m, 6H, CH_3), 1.42 (m, 1H, CH), 2.97–3.01 (m, 1H, CH-alkyl), 4.18 (m, 1H, ArCH), 5.13 (br, 1H, NH), 5.44 (br, 1H, NH), 5.57 (br, 1H, NH), 6.10 (br, 1H, NH), 7.21–7.32 (m, 2H, ArH3′H5′), 8.50–8.61 ppm (m, 2H, ArH2′H6′); elemental analysis for:
$C_{11}H_{17}N_2FPtCl_2$ (462.3): calcd: C 28.58, H 3.71, N 6.06;
(*R*,*R*)-4F-Ph/*i*Prop-PtCl_2 found: C 28.46, H 3.66, N 5.99,
(*S*,*S*)-4F-Ph/*i*Prop-PtCl_2 found: C 28.57, H 3.79, N 6.07,
(*R*,*S*)-4F-Ph/*i*Prop-PtCl_2 found: C 28.39, H 3.91, N 6.12,
(*S*,*R*)-4F-Ph/*i*Prop-PtCl_2 found: C 28.64, H 3.98, N 6.07.

Biological methods*Cell culture*: The human MCF-7 and MDA-MB 231 breast cancer cell lines, and the LNCaP/FGC cell line were obtained from the American Type Culture Collection (ATCC). Cell line banking and quality control were performed according to the seed stock concept reviewed by Hay.[19] The MCF-7 cells were maintained in L-glutamine containing Eagle's MEM (Sigma) supplemented with $NaHCO_3$ (2.2 g L^{-1}), sodium pyruvate (110 mg L^{-1}), gentamycin (50 mg L^{-1}), and 10% fetal calf serum (FCS, Gibco) using 75 cm^2 culture flasks in a humidified atmosphere (5% CO_2) at 37 °C. The MDA-MB 231 cells (McCoy's 5A medium supplemented with $NaHCO_3$ (2.2 g L^{-1}), sodium pyruvate (110 mg L^{-1}), gentamycin (50 mg L^{-1}), and 5% FCS) and the LNCaP/FGC cells (L-glutamine containing RPMI 1640 supplemented with $NaHCO_3$ (2.0 g L^{-1}), gentamycin (50 mg L^{-1}), and 7.5% FCS) were maintained under the same conditions. The cell lines were passaged weekly after treatment with trypsin (0.05%)/ethylenediaminetetraacetic acid (EDTA, 0.02%, Boehringer). Mycoplasma contamination was regularly monitored and only mycoplasma-free cultures were used.

In vitro chemosensitivity assays: The in vitro testing of the platinum complexes for antitumor activity was carried out on exponentially dividing human cancer cells according to a previously published microtiter assay.[16,20] Briefly, 100 μL of a cell suspension of 7700 cells mL^{-1} culture medium (MCF-7) resp. at 3200 cells mL^{-1} (MDA-MB 231) resp. at 2700 cells mL^{-1} (LNCaP/FGC) were plated into each well of a 96-well microtiter plate and incubated at 37 °C for 3 days resp. 5 days (LNCaP/FGC) in a humidified atmosphere

(5% CO_2). By adding an adequate volume of a stock solution of the appropriate compound (solvent: DMF) to the medium, the desired test concentration was obtained. 16 wells were used for the control (containing the appropriate amount of DMF) and each test concentration. After the proper incubation time the medium was removed, and the cells were fixed with a glutardialdehyde solution and stored under phosphate buffered saline (PBS) at 4 °C. Cell biomass was determined by a crystal violet staining technique described previously.[16,20] The efficiency of the complexes is expressed as corrected $\% T/C_{corr}$ values according to the following equations:

$$\text{Cytostatic effect: } \% T/C_{corr} = [(T - C_o)/(C - C_o)]\,100 \qquad (1)$$

$$\text{Cytocidal effect: } \% \tau = [(T - C_o)/C_o]\,100 \qquad (2)$$

in which T (test) and C (control) are the optical density values at 578 nm of the crystal violet extract of the cells in the wells (that is, the chromatin-bound crystal violet extracted with 70% ethanol), and C_o is the density of the cell extract immediately before treatment. A microplate reader at 590 nm (Flashscan AnalytikJena AG) was used for the automatic estimation of the optical density of the crystal violet extract in the wells. The calculated $\% T/C$ values can be interpreted as follows: $T/C > 80\%$: no antiproliferative effect; $80\% > T/C > 20\%$: antiproliferative effect; $20\% > T/C > 0\%$: cytostatic effect; $T/C < 0\%$: cytocidal effect

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