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Steroid Conjugates of Dichloro(6-aminomethylnicotinate)platinum(II): Effects on DNA, Sex Hormone Binding Globulin, the Estrogen Receptor, and Various Breast Cancer Cell Lines

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Esters of 6-aminomethylnicotinic acid with various steroidal alcohols were treated with K_2PtCl_4 to give the N,N-*chelated dichloroplatinum(II) complex conjugates* ***4****. Their interaction with plasmid DNA was monitored by electrophoretic mobility measurements. Their affinities towards sex hormone binding globulin (SHBG) and towards the nuclear estrogen receptor ER_α were assessed by competitive displacement radioassays. The inhibitory effect of* ***4*** *on breast tumour cells MCF-7 ER^+/ER^- and MDA-MB-231 was investigated in vitro. Conjugates with 3-O-linked estrogens* ***4a,b*** *or 17-O-linked androgens* ***4g*** *bound strongly to SHBG. The conjugate complex* ***4b****, featuring a 3-O-linked estradiol, also bound strongly and agonistically to the estrogen receptor. It also elicited distinct growth retardation of MCF-7 (ER^+) cells, presumably by a mechanism different from that of cisplatin.*

Introduction

Cisplatin still holds a salient position in the chemotherapy of various solid tumours. However, the resistance of certain tumours and occasional unwanted side effects necessitate an ongoing quest for more specific analogues.[1–5] Promising candidates are conjugates of Pt^{II} complexes with bioactive shuttle components such as porphyrins and hormones.[4–10] Sex hormones play a crucial role in the etiology and growth of the majority of breast and prostate cancers. Following the rationale that for the binding of 17β-estradiol derivatives to the nuclear estrogen receptor ER_α a free 3-OH is more important than a free 17-OH group, most Pt^{II} complex conjugates contain the steroid tethered at positions remote from the A-ring such as C16 or C17.[11–13] However, there has also been growing evidence for the involvement of sex hormone binding globulins (SHBG) in the development of sex-hormone-dependent tumours. SHBG acts as a carrier for estrogens and androgens and also interacts with plasma membranes of cells in a ligand-dependent manner, thereby stimulating intracellular signalling pathways that alter cell growth and function.[14] Interestingly, estrogens slip into the SHBG host D-ring first, which would require a 3-O rather than a 17-O linkage in the corresponding estradiol–Pt^{II} complex conjugates.

Herein we report a short synthesis of new dichloro(6-aminomethylnicotinate)platinum(II) complexes[13,15] linked by ester bonds to either 3-O or 17-O of various steroids commonly associated with binding to transport proteins and nuclear receptors. Their binding affinities to human wild-type SHBG and to the nuclear estrogen receptor were ascertained, as were their cytotoxicities against hormone-dependent and independent breast tumour cell types in vitro.

Chemistry

The platinum complexes **4** were obtained by reaction of the preformed ligands, the chlorides or trifluoroacetates of 6-ammoniummethylnicotinates **3**, with K_2PtCl_4 (Scheme 1). Initially, the N-Boc-protected 6-aminomethylnicotinic acid **1**[16] was esterified under Yamaguchi conditions with the appropriate steroidal alcohol to afford the N-Boc-protected nicotinates **2**. These were deprotected with 4 M HCl/dioxane to give the ammonium chlorides, or with 50% trifluoroacetic acid (TFA) in dichloromethane to give the ammonium trifluoroacetates **3**. In

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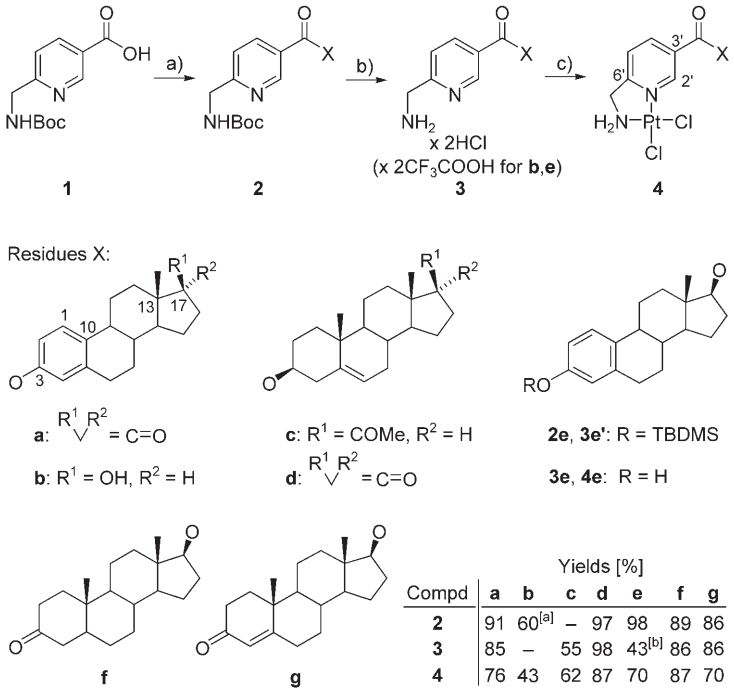
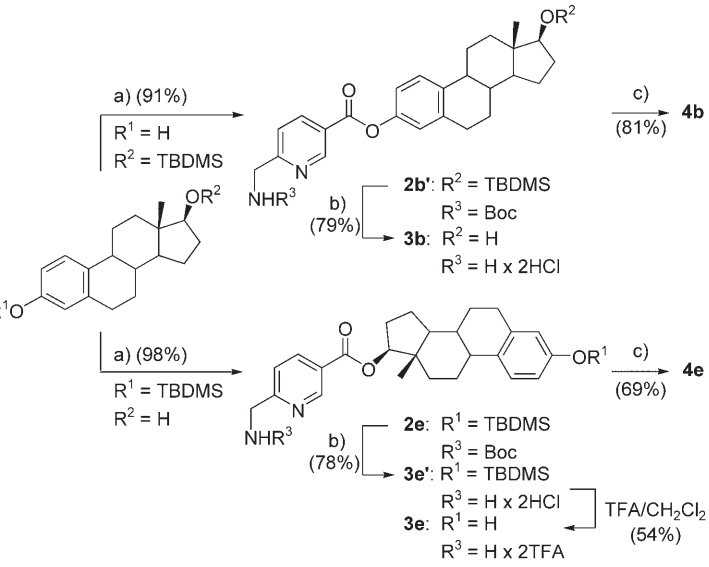


Compd	Yields [%] a	b	c	d	e	f	g
2	91	60[a]	–	97	98	89	86
3	85	–	55	98	43[b]	86	86
4	76	43	62	87	70	87	70

Scheme 1. Reagents and conditions: a) Et_3N, $C_6H_2Cl_3COCl$, 4-(dimethylamino)pyridine (DMAP), steroid alcohol, DMF/toluene, RT; [a] **2b** from **2a**: $NaBH_4$, THF, RT, 24 h. b) 4 M HCl/dioxane, RT, 1 h (for **a**,**c**–**g**); TFA/CH_2Cl_2 (1:1), 1 h (for **b**). c) K_2PtCl_4, H_2O/THF, RT, 24 h; [b] over two steps via **3e**′. (– in yields = not isolated).

this way, estrone (run **a**), pregnenolone (run **c**), and dehydro-*epi*-androsterone (run **d**) were attached through the oxygen atom at C3 to the stationary chelate ligand, while dihydrotestosterone (run **f**) and testosterone (run **g**) were esterified at 17-O. Estradiol was linked either way (runs **b** and **e**). Its 3-O ester **2b** was prepared by the reduction of estrone ester **2a** with $NaBH_4$. Alternatively, 6-ammoniummethylnicotinate **3b** was prepared from 17-O-*tert*-butyldimethylsilyl (TBDMS)-protected estradiol via bisprotected 3-O ester **2b**′ (Scheme 2). The 17-O ester **3e** of estradiol was obtained starting from 3-O-TBDMS-protected estradiol to give nicotinate **2e**, which was converted into the 6-ammoniummethylnicotinate **3e**′. Desilylation of 3-O with TFA/CH_2Cl_2 left the bistrifluoroacetate **3e**. Finally, reaction of compounds **3** with K_2PtCl_4 in mixtures of water and tetrahydrofuran (THF) at room temperature under slightly acidic conditions afforded complexes **4**. Their ^{1}H NMR spectra in [D_7]DMF showed a conspicuous downfield shift for the 2-H protons (e.g. $\Delta\delta = -0.86$ ppm for the couple **3a**/**4a**) and ^{195}Pt satellites for the 2-H and the exocyclic NH_2 and CH_2 protons of the aminomethylnicotinate. The ^{195}Pt NMR signals lay between 2439–2441 ppm, and had line widths of 250–300 Hz, which is typical of N-ligated platinum.



Scheme 2. Reagents and conditions: a) **1**, Et_3N, $C_6H_2Cl_3COCl$, DMAP, DMF/toluene, RT, 24 h. b) 4 M HCl/dioxane, RT, 1 h. c) K_2PtCl_4, H_2O/THF, RT, 24 h.

Interaction with Sex Hormone Binding Globulin (SHBG)

SHBG is a human plasma glycoprotein produced by the liver and certain tissues, and acts as a specific carrier and in the storage of sex steroids.[17] It also inhibits the estradiol-induced proliferation of certain breast cancer cells by reversing the anti-apoptotic effect of the latter when binding to specific sites in the membrane.[18] From crystal structure data it is known that SHBG binds androgen and estrogen molecules in opposite orientations. While androgens slip into the binding cavity of the host with the A-ring first, estrogens go in D-ring first.[17] As any shuttle effect of steroids in drug conjugates depends on their recognition and binding by their biocarriers, we measured the binding affinities of complexes **4** towards SHBG by competitive displacement of [^{3}H]DHT (dihydrotestosterone) from the human SHBG steroid binding site. Figure 1 depicts the concentration response curves for estradiol, testosterone, and all complexes **4** that were recognised and bound by SHBG. The relative binding affinities (RBA) quoted in the legend are defined as the ratio of the concentrations of steroid competitors resulting in a 50% reduction in specific binding of [^{3}H]DHT to the concentration of testosterone required to produce the same 50% reduction effect. Clearly, complex **4b**, which features a 3-O-bound estradiol, binds strongly to SHBG (IC_{50} = 470 nM, RBA = 29) very likely in the same orientation as the free steroid, whereas the complex **4e**, with a 17-O bound estradiol, does not bind at all. Complex **4a**, containing a 3-O-bound estrone, was also a good ligand for SHBG (IC_{50} = 1230 nM, RBA = 72). The complexes **4f** and **4g**, containing 17-O-bound androgens, were also recognised by SHBG, however testosterone complex **4g** (IC_{50} = 2780 nM, RBA = 163) markedly more so than dihydrotestosterone complex **4f**. The complexes **4c** and **4d** proved to be poor binders.

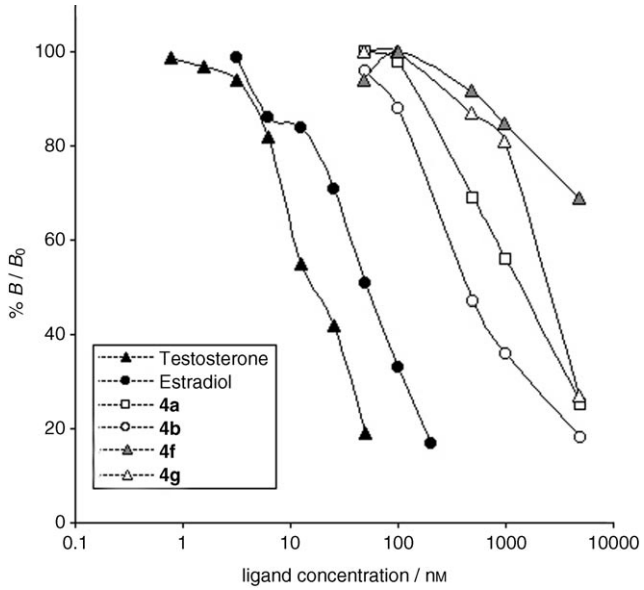


Figure 1. Competitive displacement of [^{3}H]DHT from the human SHBG steroid binding site by testosterone, estradiol, and various complexes **4**. *B* and B_0 are the amounts of the [^{3}H]DHT tracer specifically bound to human SHBG in the presence and absence of a steroid competitor, respectively. Relative binding affinities (RBA) as defined in the text: testosterone = 1, estradiol = 4, **4a** = 72, **4b** = 29, **4g** = 163.

Interaction with DNA

Solutions of complexes **4** in DMSO were incubated at different concentrations with pBR322 plasmid DNA, and the effect on the electrophoretic mobilities of the various DNA forms present was monitored. A strong retardation of the front-most covalently closed circular (*ccc*) form was observed for **4e** and **4f** due to an unwinding of the supercoiled DNA by platinum adduct formation. At a concentration of 60 μM, the open circular (*oc*) and *ccc* forms gave rise to a single coalescing band. Generally, the bands tend to get darker towards higher concentrations of **4** as the staining agent ethidium bromide can intercalate less effectively into the platinum-laden unwound DNA (Figure 2). This effect is in line with a linear decrease of the fluorescence intensity of DNA–ethidium bromide adducts when plotted against increasing concentrations of **4e** or **4f**

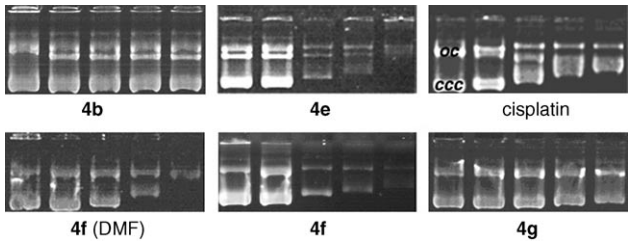


Figure 2. Modification of gel electrophoretic mobility of pBR322 plasmid DNA when incubated with different concentrations of DMSO solutions of selected complexes **4** and of cisplatin (*oc* = open circular, *ccc* = covalently closed circular DNA form). For comparison, the effect of compound **4f** dissolved in DMF is also shown. Concentrations (in μM): lane 1 (leftmost): 0, lane 2: 5, lane 3: 20, lane 4: 40, lane 5: 60.

added to plasmid or salmon sperm DNA. Complexes **4d** and **4g** also caused retardation of the *ccc* form, albeit starting only at concentrations well above 50 μM. Virtually no effect was visible in assays with complexes **4a**, **4b**, **4c**, or with the free steroids, or with dichloro(6-aminomethylnicotinic acid)platinum(II), a conceivable hydrolysis product of complexes **4**. These compounds either had a low binding affinity or their binding did not change the degree of DNA superhelicity. The unwinding effect of cisplatin is also shown in Figure 2 for comparison, as is the effect of compound **4f** when applied as a DMF solution. Generally, solutions of complexes **4** in DMF are more stable than those in DMSO due to a slower displacement of the chloride ligands. Their impact on DNA thus sets on more slowly but reaches a similar terminal magnitude. The pronounced differences in the electrophoretograms with complexes that have similar steroid structures are remarkable. For instance, the weak effector **4b** carries a 3-O-linked estradiol, and the strong effector **4e**, a 17-O-linked estradiol. Here, the different location of the aromatic π system and of the puckered cycloalkanes might allow different interactions with the neighbouring DNA bases and the phosphate backbone. Likewise, the more strongly puckered, bulkier tetracycle of dihydrotestosterone might be responsible for the greater distorting effect of its conjugate complex **4f** in comparison with the flatter testosterone moiety in **4g**.

Cytotoxicities and Receptor Affinities

The toxicities of complexes **4** were investigated in vitro against cell lines of the hormone-dependent breast cancer MCF-7 (ER^+) and the hormone-independent breast cancers MCF-7 (ER^-) and MDA-MB-231, and compared with those of cisplatin and of the estrogen receptor modulator 4-hydroxytamoxifen (Figure 3 and Supporting Information). Estradiol has been shown to promote the growth of MCF-7 (ER^+) breast cancer cells at low concentrations by interacting with its nuclear receptor to induce the production of growth factors[19] and by preventing apoptosis.[20] Osella and co-workers had surmised that the overall proliferative effect of their *cis*-[Pt(en)Cl_2]–estradiol conjugates might be due to the proliferative effect of the estrogenic component overriding its biocarrier effect and any potential DNA damage by the platinum complex moiety.[21] Contrary to that, our 3-O-linked estradiol conjugate **4b** exhibited a distinct inhibitory effect on the growth of MCF-7 (ER^+) cells with an IC_{50} value of ~15–20 μM over long-term (259 h) treatments. We made sure that potential hydrolysis products of **4b** are not involved. Complex **4** (X = OH) with a free carboxylic acid was inactive in MCF-7 cells, while free estradiol was cytocidal at concentrations above 15 μM (see Supporting Information). As expected, all compounds **4** had little if any effect on MDA-MB-231 and MCF-7 (ER^-) cells at concentrations below 20 μM. Cisplatin and hydroxytamoxifen were also less effective towards these two cell lines than towards MCF-7 (ER^+).[22] Interestingly, the addition of murine sera enriched in human SHBG (20 nM effective physiological-like concentration) to MCF-7 (ER^+) cells either simultaneously with, prior to, or following treatment with complex **4b** (10 μM) did not significantly

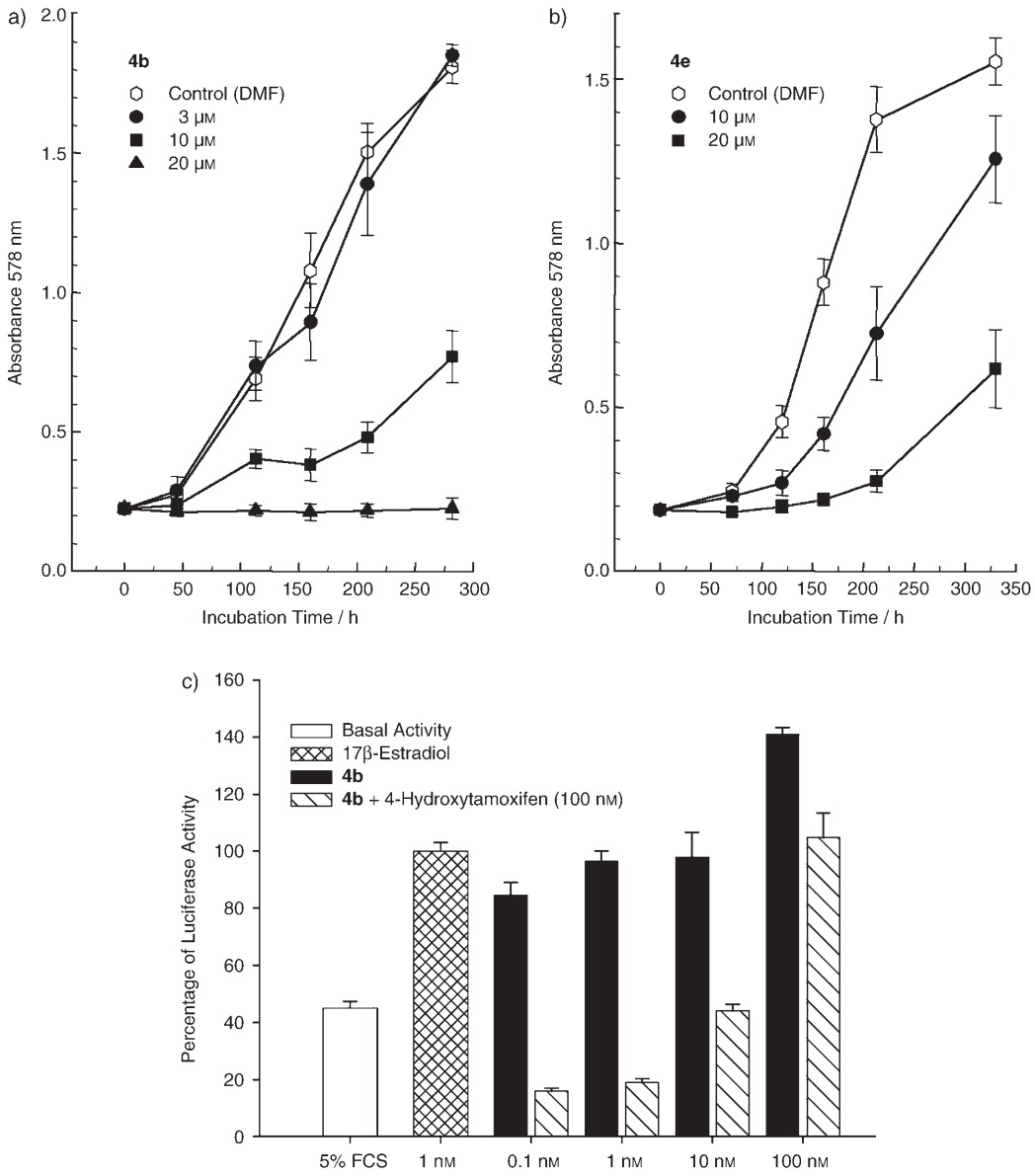


Figure 3. Growth inhibitory effect in long-term treatment of the human hormone-sensitive breast cancer cell line MCF-7 (HTB 22) in passage 169 from origin, with various concentrations of conjugate complexes a) **4b** and b) **4e** (added as solutions in DMF), as assessed by crystal violet staining. c) Estrogenic activity of **4b** and mixtures of **4b** and 4-hydroxytamoxifen relative to that of 17β-estradiol, as assessed by luciferase gene reporter assay.

change its growth inhibitory effect (see Supporting Information). Fortunati and co-workers found that pre-incubation of MCF-7 (ER^+) cells with SHBG before estradiol treatment counteracted the anti-apoptotic effect of the latter.[18a]

To prove that compound **4b** is still recognised by the nuclear estrogen receptor, its binding affinity was measured by competitive displacement of specifically bound [2,4,6,7-^{3}H]17β-estradiol from calf uterine cytosol as the source of ER_α. The binding constant K_i as calculated according to the Cheng–Prusoff equation was found to be 27.5 ± 5.5 nM. This means a rather strong binding, which is still sevenfold weaker than that of 4-hydroxytamoxifen, with $K_i = 3.8$ nM.[23] Whereas isomer **4e**, which bears a 17-O-linked estradiol, is a stronger binder to plasmid DNA than **4b** and does not bind SHBG, bound less effectively to the receptor ER_α ($K_i = 36 \pm 16$ nM) and also inhibited MCF-7 (ER^+) tumour cells to a lesser, though still notable, extent (Figure 3). No affinity to the ER was detected for compound **4f** up to a concentration of 10 μM, whereas 10 μM **4g** displaced the specifically bound [^{3}H]17β-estradiol from the ER by approximately 35%. A luciferase gene reporter assay[23] revealed a strong concentration-dependent estrogenic (agonistic) effect upon binding of **4b** similar to that of free 17β-estradiol. As shown in Figure 3, the stimulatory effect of **4b** was antagonized by the anti-estrogen 4-hydroxytamoxifen.

To exclude a conceivable involvement of free steroids in receptor binding by way of hydrolysis of conjugates **4**, 2 mM solutions of the latter in aqueous buffers (PBS/DMF 9:1 *v/v*, pH 7.5) were incubated at 37 °C for 2 days. Subsequent HPLC analysis (column: Ultrasep ES 100 RP18; eluent: methanol/water 70:30 *v/v*) revealed the integrity of complexes **4** and the

absence of more than trace amounts of the free steroid alcohols (see Supporting Information).

Conclusions

As a proof of principle, we have shown that conjugates of appropriate platinum complexes with estrogens attached at the 3-OH group are still recognised by the biocarrier SHBG and by the nuclear estrogen receptor. This is a prerequisite for any active transport to, and directed accumulation at the target site. We could also show that the docking of the conjugate at the receptor is nondestructive, not modulated by SHBG, and still provokes a strong concentration-dependent estrogenic response. Compound **4b**, the most active in these and in the cytotoxicity tests, exerted no DNA distorting effect, suggesting a mode of action different from that of cisplatin. The net inhibiting effect of conjugate complex **4b** on the growth of MCF-7 (ER^+) also leaves room for further enhancement by altering structural features such as the stationary diamino ligand or the spacer between complex and steroid portions, or by replacing the estrogenic steroids with antagonistic receptor binders.

Experimental Section

Methods: 1. Interaction with pBR322 plasmid DNA: Interaction of complexes **4** with pBR322 plasmid DNA was studied by agarose gel electrophoresis following the general method by Huq and coworkers.[24] Briefly, aliquots (20 μL) of a stock TE buffer solution of pBR322 DNA (73.5 $\mu g\,mL^{-1}$) were incubated in the presence of increasing concentrations of compounds **4** (0, 5, 20, 40, and 60 μM). Incubation was carried out in the dark at 37 °C for 24 h. Each assay was treated with 2 μL loading buffer (6 mL glycerol, 20 mg xylene cyanol, 1 mL 5×TBE (Tris–borate–EDTA) buffer, 3 mL H_2O); 11 μL of the resulting mixture containing 0.735 μg plasmid DNA was loaded onto a 1% agarose gel, and electrophoresis was carried out in a 0.5×TBE buffer (0.045 M Tris-base, 1 mM EDTA, 0.045 M boric acid) for 4 h at 66 V. At the end of electrophoresis, the gel was stained in the same buffer containing ethidium bromide (0.5 $\mu g\,mL^{-1}$) and visualised under UV light.

2. Cytotoxicity assay: *Cell line and culture conditions:* The human estrogen-receptor-positive MCF-7 (HTB 22) and the estrogen-receptor-negative MDA-MB-231 (HTB 26) breast cancer cells were obtained from the American Type Culture Collection (ATCC), Rockville, MD (USA), whereas the estrogen-receptor-negative variant MCF-7 (ER^-) was established from wild-type cells, which had spontaneously lost the receptor as demonstrated previously.[25] Cell banking and quality control were performed according to the "seed stock concept". To avoid potential differential inactivation of the platinum complexes by reaction with components of the culture media, all breast cancer cells were cultured in Eagle's minimum essential medium (Sigma, Munich, Germany) containing L-glutamine, $NaHCO_3$ (2.2 $g\,L^{-1}$), sodium pyruvate (110 $mg\,L^{-1}$), and 5% fetal calf serum (FCS; Biochrom, Berlin, Germany). Cells were maintained under a water-saturated atmosphere (5% CO_2) at 37 °C in 75-mL culture flasks (Nunc, Wiesbaden, Germany), and were serially passaged following trypsinisation by using 0.05% trypsin/0.02% EDTA (PAA laboratories, Cölbe, Germany). Mycoplasma contamination was routinely monitored, and only mycoplasma-free cultures were used.

Test compounds: Cisplatin and 4-hydroxytamoxifen were purchased from Sigma (Munich, Germany) and used as 10 mM stock solutions in DMF stored at −20 °C.

Chemosensitivity assay: Assays were performed as described previously.[25] In brief, tumour cell suspensions (~4×10^6 $cells\,L^{-1}$) were seeded (100 $\mu L\,well^{-1}$) into 96-well flat-bottomed microtitration plates (Greiner, Frickenhausen, Germany). After 2–3 days, the culture medium was removed by suction and replaced by fresh medium (200 $\mu L\,well^{-1}$) containing varying drug concentrations or vehicle (DMF). Drugs were added by a 1000-fold dilution of the appropriate concentrated solutions with culture medium. On every plate, 16 wells served as controls and 16 wells were used per drug concentration. After various times of incubation, the cells were fixed with glutardialdehyde (Merck, Darmstadt, Germany) and stored in a refrigerator. At the end of the experiment all plates were processed simultaneously by staining with a solution of aqueous crystal violet (0.02%, 100 $\mu L\,well^{-1}$; Merck, Darmstadt, Germany). Excess dye was removed by rinsing the trays with water for 20 min. The stain bound by the cells was redissolved in 70% ethanol (200 $\mu L\,well^{-1}$) while shaking the microplates for about 3 h. Absorbance (proportional to cell mass) was measured at 578 nm using a BIOTEK 309 Autoreader (Tecnomara, Fernwald, Germany). The results (mean values ± SD) were plotted as growth curves.

3. SHBG ligand binding assay: An established competitive ligand binding assay was used to determine the binding affinities of complexes **4** to human SHBG in relation to those of testosterone and estradiol.[26] In brief, the assay involved mixing aliquots (100 μL) of diluted (1:200) human pregnancy serum containing approximately 1 nM SHBG, which was pretreated with dextran-coated charcoal (DCC) to remove endogenous steroid ligand, with 100 μL of tritium-labelled DHT ([^{3}H]DHT) at 10 nM as the labelled ligand. For the screening assay, triplicate aliquots (100 μL) of a fixed amount (200 μL) of test compound were added to this SHBG/[^{3}H]DHT mixture and incubated overnight at room temperature, followed by 10 min incubation with a DCC slurry (500 μL) at 0 °C and centrifugation to separate SHBG-bound from free [^{3}H]DHT. Compounds that displaced more than 35% of the [^{3}H]DHT from the SHBG in this assay were then diluted serially, and triplicate aliquots (100 μL) of known concentrations of test compounds were used to generate complete competition curves by incubation with the SHBG/[^{3}H]DHT mixture, and separation of SHBG-bound from free [^{3}H]DHT, as in the screening assay. The amounts of [^{3}H]DHT bound to SHBG at each concentration of competitor ligand were determined by scintillation spectrophotometry and plotted in relation to the amount of [^{3}H]DHT bound to SHBG at zero concentration of competitor. From the resulting competition curves, IC_{50} values and relative binding affinity parameters (RBA) could be calculated if displacement of more than 50% of [^{3}H]DHT from SHBG was achieved.

4. Estrogen receptor ligand binding assay: The affinity to the estrogen receptor was measured in a competitive radioligand assay[23] using calf uterus cytosol as receptor source and 1 nM [2,4,6,7-^{3}H]17β-estradiol (specific activity: 3.48 $TBq\,mmol^{-1}$; Amersham Biosciences Europe GmbH, Freiburg, Germany) as tracer. Freshly excised calf uteri were freed from fat, perimetrium, and parametrium, washed with a solution of 0.9% NaCl, and cut into small pieces. One mL TED–Mo buffer (10 mM Tris, 10 mM sodium molybdate, 1 mM EDTA, 0.5 mM DTE, pH 7.4, supplemented with Complete (EDTA-free protease inhibitor, Roche Diagnostics, Mannheim, Germany; one tablet per 50 mL) was added per gram of tissue. The suspension was homogenized (three to five times for ~10 s) with an Ultraturrax and centrifuged at 10500 *g* for 90 min. Before storage at −78 °C, the protein concentration of the clear cy-

tosol (~20 mg mL^{-1}) was determined according to the Bradford method. For the ER ligand binding assay, the cytosol was diluted with Tris buffer (pH 7.5) to a protein concentration of 5 mg mL^{-1}.

The incubation mixture (500 μL) included 1 nM [^{3}H]17β-estradiol (added in 100 μL Tris buffer, 0.01 M, pH 7.5 supplemented with EDTA (0.01 M) and NaN_3 (0.003 M)), 10^{-10}–10^{-6} M competing ligand (in 100 μL buffer), 100 μL of calf uterine cytosol, and buffer. The mixture was incubated for 18–24 h at 4 °C, after which 0.5 mL DCC slurry (0.8% charcoal Norit A (Serva, Heidelberg) and 0.008% dextran in buffer) was added to the tubes, and the contents were mixed. The tubes were incubated for 90 min at 4 °C and then centrifuged at 2500 *g* for 10 min to pellet the charcoal. An aliquot (100 μL) of the supernatant was removed, and the radioactivity was determined by an LS 6500 Beckmann–Coulter scintillation counter after addition of 3 mL Rotiszint eco plus (Roth, Karlsruhe, Germany). Nonspecific binding was calculated using 17β-estradiol (4 μM) as competing ligand.

For the quantitation of estrogen receptor binding, K_i values were calculated according to the Cheng–Prusoff equation (K_d for [2,4,6,7-^{3}H]17β-estradiol = 0.60 nM).[27] IC_{50} values were determined with SigmaPlot 9.0 by logit–log plots of the data from three independent experiments.

5. Luciferase gene reporter assay: MCF-7/2a cells with the reporter construct integrated in the genome were cloned and characterised as described elsewhere.[28] The MCF-7/2a cells were maintained in Dulbecco's modified Eagle's medium without phenol red (Gibco, Karlsruhe, Germany), supplemented with L-glutamine (292 mg L^{-1}), 175 mg L^{-1} G-418 (PAA Laboratories, Cölbe, Germany) and 5% FCS. Determination of the estrogenic activity was essentially performed as described,[23] with the following modifications: dextran/charcoal-treated fetal calf serum was replaced with FCS, luciferase activity was quantified with the firefly luciferase kit (Biotrend, Köln, Germany) according to the manufacturer's instructions (Biotium Inc., Hayward, CA, USA).

Syntheses: 1. Syntheses of N-Boc-protected 6-aminomethylnicotinates (2): *6′-(tert-Butoxycarbonylaminomethyl)nicotinic acid 17-oxoestra-1,3,5(10)-trien-3-yl ester (**2a**):* 6-*tert*-Butoxycarbonylaminomethylnicotinic acid **1**[16] (160 mg, 0.63 mmol) was dissolved in DMF (2 mL) and treated with Et_3N (100 μL, 0.72 mmol) and 2,4,6-trichlorobenzoyl chloride (111 μL, 0.72 mmol). The resulting suspension was stirred at room temperature for 20 min. A solution of estrone (341 mg, 1.26 mmol) and DMAP (155 mg, 1.26 mmol) in toluene (20 mL) was added, and the reaction mixture was stirred at room temperature for 16 h. After dilution with ethyl acetate and washing with water, the organic phase was dried over Na_2SO_4 and concentrated. The residue was purified by column chromatography (silica gel 60; ethyl acetate/*n*-hexane 1:2). Yield: 290 mg (0.57 mmol, 91%); colourless oil; R_f = 0.16 (ethyl acetate/*n*-hexane 1:2); ν_{max} (ATR): $\tilde{\nu}$ = 3297, 2974, 2931, 1735, 1700, 1598, 1493, 1258, 1163, 1080 cm^{-1}; ^{1}H NMR (300 MHz, $CDCl_3$): δ = 0.91 (3H, s, 13-Me), 1.45 (9H, s, CMe_3), 1.5–2.6 (16H, m, steroid-H), 2.9–3.0 (2H, m, 6-H), 4.52 (2H, d, ^3J = 5.6 Hz, CH_2N), 5.5–5.6 (1H, m, NH), 6.9–7.0 (2H, m, 4-H, 2-H), 7.33 (1H, d, ^3J = 8.3 Hz, 1-H), 7.41 (1H, d, ^3J = 8.2 Hz, 5′-H), 8.38 (1H, d, ^3J = 8.2 Hz, 4′-H), 9.26 ppm (1H, s, 2′-H); ^{13}C NMR (75.5 MHz, $CDCl_3$): δ = 13.8 (13-Me), 21.6 (C-15), 25.8 (C-11), 26.3 (C-7), 28.4 (*CMe*$_3$), 29.4 (C-6), 31.5 (C-12), 35.8 (C-16), 38.0 (C-8), 44.2 (C-9), 45.9 (C-13), 47.9 (CH_2N), 50.4 (C-14), 78.8 (*C*Me$_3$), 118.7 (C-2), 121.2 (C-4), 121.5 (C-5′), 124.3 (C-3′), 126.6 (C-1), 137.8 (C-5), 138.2 (C-4′), 138.2 (C-10), 148.4 (C-3), 150.9 (C-2′), 156.4 (OCON), 162.7 (C-6′), 164.0 (CO_2), 209.7 ppm (CO); *m/z* (EI) 504 (4) [M^+], 430 (38), 269 (17), 235 (11), 179 (99), 161 (100), 135 (61); accurate mass (EIMS) for $C_{30}H_{36}N_2O_5$: calcd 504.26242, obsd 504.26240.

*6′-(tert-Butoxycarbonylaminomethyl)nicotinic acid 17-hydroxyestra-1,3,5(10)-trien-3-yl ester (**2b**):* A solution of **2a** (100 mg, 0.2 mmol) in THF (5 mL) was treated with $NaBH_4$ (20 mg, 0.53 mmol). The reaction mixture was stirred at room temperature for 16 h. Water was added, and the mixture was extracted with ethyl acetate. The organic phase was dried over Na_2SO_4, concentrated, and the residue was purified by column chromatography (silica gel 60; ethyl acetate/hexane 1:1). Yield: 60 mg (0.12 mmol, 60%); colourless oil; R_f = 0.27 (ethyl acetate/*n*-hexane 1:1); ν_{max} (ATR): $\tilde{\nu}$ = 3363, 2930, 2869, 1737, 1695, 1600, 1493, 1248, 1150, 906, 727 cm^{-1}; ^{1}H NMR (300 MHz, $CDCl_3$): δ = 0.77 (3H, s, 13-Me), 1.1–2.3 (22H, m, CMe_3, steroid-H), 2.8–2.9 (2H, m, 6-H), 3.6–3.8 (1H, m, 17-H), 4.51 (2H, d, ^3J = 5.6 Hz, CH_2N), 6.6–6.7 (1H, m, NH), 6.8–7.0 (2H, m, 2-H, 4-H), 7.32 (1H, d, ^3J = 8.4 Hz, 1-H), 7.41 (1H, d, ^3J = 8.2 Hz, 5′-H), 8.38 (1H, d, ^3J = 8.2 Hz, 4′-H), 9.26 ppm (1H, s, 2′-H); ^{13}C NMR (75.5 MHz, $CDCl_3$): δ = 11.0 (13-Me), 23.1 (C-15), 26.1 (C-11), 27.0 (C-7), 28.4 (*CMe*$_3$), 29.5 (C-6), 30.5 (C-16), 36.6 (C-12), 38.4 (C-8), 43.2 (C-13), 44.1 (C-9), 45.9 (CH_2N), 50.1 (C-14), 79.9 (*C*Me$_3$), 81.8 (C-17), 118.5 (C-2), 121.4 (C-4, C-5′), 124.4 (C-3′), 126.5 (C-1), 138.3 (C-4′), 138.4 (C-5), 138.5 (C-10), 148.2 (C-3), 150.8 (C-2′), 156.0 (OCON), 162.7 (C-6′), 164.0 ppm (CO_2); *m/z* (EI) 506 (6) [M^+], 450 (13), 433 (10), 406 (18), 271 (25), 179 (60), 135 (100); $C_{30}H_{38}N_2O_5$ requires: C 71.12, H 7.56, N 5.53; found: C 71.18, H 7.41, N 5.43.

*6′-(tert-Butoxycarbonylaminomethyl)nicotinic acid 17β-(tert-butyldimethylsilyloxy)estra-1,3,5(10)-trien-3-yl ester (**2b′**):* Analogously to the synthesis of **2a**, compound **2b′** (310 mg, 0.50 mmol, 91%) was obtained from **1** (140 mg, 0.56 mmol), Et_3N (100 μL, 0.72 mmol), 2,4,6-trichlorobenzoyl chloride (111 μL, 0.72 mmol), 17β-TBDMS-estradiol (230 mg, 0.60 mmol), obtained from the reaction of β-estradiol and TBDMSCl,[29] and DMAP (155 mg, 1.26 mmol) as a colourless solid of mp: 150 °C; R_f = 0.22 (ethyl acetate/*n*-hexane 1:4); ν_{max} (ATR): $\tilde{\nu}$ = 3326, 2928, 1735, 1701, 1600, 1495, 1248, 1141, 1085, 1019, 883, 774 cm^{-1}; ^{1}H NMR (300 MHz, $CDCl_3$): δ = 0.01 (3H, s, SiMe), 0.02 (3H, s, SiMe), 0.74 (3H, s, 13-Me), 0.88 (9H, s, $SiCMe_3$), 1.1–2.4 (22H, m, $OCMe_3$, steroid-H), 2.8–2.9 (2H, m, 6-H), 3.63 (1H, t, ^3J = 8.2 Hz, 17-H), 4.52 (2H, d, ^3J = 5.5 Hz, CH_2N), 5.5–5.6 (1H, m, NH), 6.90 (1H, s, 4-H), 6.95 (1H, d, ^3J = 8.5 Hz, 2-H), 7.32 (1H, d, ^3J = 8.5 Hz, 1-H), 7.40 (1H, d, ^3J = 8.2 Hz, 5′-H), 8.38 (1H, d, ^3J = 8.2 Hz, 4′-H), 9.27 ppm (1H, s, 2′-H); ^{13}C NMR (75.5 MHz, $CDCl_3$): δ = −4.8 (SiMe), −4.5 (SiMe), 11.4 (13-Me), 18.1 (Si*C*Me$_3$), 23.3 (C-15), 25.9 (Si*CMe*$_3$), 26.3 (C-11), 27.1 (C-7), 28.4 (O*CMe*$_3$), 29.6 (C-6), 31.0 (C-16), 37.1 (C-12), 38.5 (C-8), 43.6 (C-13), 44.3 (C-9), 45.9 (CH_2N), 49.7 (C-14), 79.3 (*C*Me$_3$), 81.7 (C-17), 118.4 (C-2), 121.2 (C-4), 121.4 (C-5′), 124.4 (C-3′), 126.6 (C-1), 138.2 (C-4′), 138.6 (C-5), 138.7 (C-10), 148.2 (C-3), 150.9 (C-2′), 156.0 (OCON), 162.6 (C-6′), 164.0 ppm (CO_2); *m/z* (EI) 620 (7) [M^+], 507 (82), 463 (38), 387 (17), 179 (25), 135 (100); accurate mass (EIMS) for $C_{36}H_{52}N_2O_5Si$: calcd 620.36455, obsd 620.36460.

*6′-(tert-Butoxycarbonylaminomethyl)nicotinic acid 17-oxoandrost-5-en-3β-yl ester (**2d**):* As with **2a**, compound **2d** (310 mg, 0.59 mmol, 97%) was prepared from **1** (160 mg, 0.63 mmol), Et_3N (100 μL, 0.72 mmol), 2,4,6-trichlorobenzoyl chloride (111 μL, 0.72 mmol), dehydroandrosterone (366 mg, 1.26 mmol), and DMAP (155 mg, 1.26 mmol) as a colourless solid of mp: 180 °C; R_f = 0.56 (ethyl acetate/*n*-hexane 1:1); ν_{max} (ATR): $\tilde{\nu}$ = 2941, 1726, 1707, 1596, 1520, 1298, 1254, 1163, 1122, 1022 cm^{-1}; ^{1}H NMR (300 MHz, $CDCl_3$): δ = 0.88 (3H, s, 13-Me), 1.0–2.6 (31H, m, CMe_3, steroid-H), 4.48 (2H, d, ^3J = 5.5 Hz, CH_2N), 4.8–5.0 (1H, m, 3-H), 5.4–5.6 (2H, m, 6-H, NH), 7.33 (1H, d, ^3J = 8.2 Hz, 5′-H), 8.23 (1H, d, ^3J = 8.2 Hz, 4′-H), 9.10 ppm (1H, s, 2′-H); ^{13}C NMR (75.5 MHz, $CDCl_3$): δ = 13.5 (13-Me), 19.4 (10-Me), 20.3 (C-11), 21.9 (C-15), 27.8 (C-2), 28.4 (*CMe*$_3$), 30.8 (C-12), 31.4 (C-7), 31.5 (C-8), 35.8 (C-16), 36.8 (C-10), 36.9 (C-4), 38.1 (C-1), 45.8 (CH_2N), 47.5 (C-13), 50.2 (C-9), 51.7 (C-14), 74.9 (C-3), 79.8

(CMe_3), 121.0 (C-5′), 122.2 (C-6), 125.2 (C-3′), 137.7 (C-4′), 139.7 (C-5), 150.4 (C-2′), 155.9 (OCON), 161.9 (C-6′), 164.6 (CO_2), 221.0 ppm (CO); *m/z* (EI) 523 (1) [MH^+], 449 (10), 270 (100), 255 (16), 197 (20), 179 (20), 121 (25); $C_{31}H_{42}N_2O_5$ requires: C 71.24, H 8.10, N 5.36; found: C 71.36, H 8.32, N 5.30.

*6′-(tert-Butoxycarbonylaminomethyl)nicotinic acid 3-(tert-butyldimethylsilyloxy)estra-1,3,5(10)-trien-17β-yl ester (**2e**):* As with **2a**, compound **2e** (340 mg, 0.55 mmol, 98%) was prepared from **1** (140 mg, 0.56 mmol), Et_3N (100 μL, 0.72 mmol), 2,4,6-trichlorobenzoyl chloride (111 μL, 0.72 mmol), 3β-TBDMS-estradiol (230 mg, 0.60 mmol), and DMAP (155 mg, 1.26 mmol) as a colourless solid of mp: 130 °C; R_f = 0.18 (ethyl acetate/*n*-hexane 1:4); ν_{max} (ATR): $\tilde{\nu}$ = 3273, 2929, 2856, 1710, 1600, 1495, 1287, 1123, 838 cm^{-1}; ^1H NMR (300 MHz, $CDCl_3$): δ = 0.15 (6H, s, SiMe), 0.9–1.0 (12H, m, 13-Me, $SiCMe_3$), 1.3–2.4 (22H, m, $OCMe_3$, steroid-H), 2.8–2.9 (2H, m, 6-H), 4.48 (2H, d, ^3J = 5.5 Hz, CH_2N), 4.93 (1H, t, ^3J = 8.4 Hz, 17-H), 5.5–5.6 (1H, m, NH), 6.54 (1H, s, 4-H), 6.59 (1H, d, ^3J = 8.4 Hz, 2-H), 7.10 (1H, d, ^3J = 8.4 Hz, 1-H), 7.34 (1H, d, ^3J = 8.1 Hz, 5′-H), 8.24 (1H, d, ^3J = 8.1 Hz, 4′-H), 9.13 ppm (1H, s, 2′-H); ^{13}C NMR (75.5 MHz, $CDCl_3$): δ = −4.4 (SiMe), 12.4 (13-Me), 18.2 ($SiCMe_3$), 23.4 (C-15), 25.7 ($SiCMe_3$), 26.1 (C-11), 27.3 (C-7), 27.8 (C-16), 28.4 ($OCMe_3$), 29.6 (C-6), 37.0 (C-12), 38.5 (C-8), 43.4 (C-13), 43.8 (C-9), 45.8 (CH_2N), 49.9 (C-14), 79.7 (CMe_3), 83.8 (C-17), 117.2 (C-2), 120.0 (C-4), 121.1 (C-5′), 125.2 (C-3′), 126.1 (C-1), 132.8 (C-10), 137.7 (C-4′, C-5), 150.4 (C-2′), 153.4 (C-3), 159.4 (OCON), 161.9 (C-6′), 165.1 ppm (CO_2); *m/z* (EI) 620 (74) [M^+], 546 (96), 520 (82), 489 (60), 463 (91), 253 (86), 209 (100), 178 (45), 151 (82), 135 (83); accurate mass (EIMS) for $C_{36}H_{52}N_2O_5Si$: calcd 620.36455, obsd 620.36460.

*6′-(tert-Butoxycarbonylaminomethyl)nicotinic acid 3-oxoandrostan-17β-yl ester (**2f**):* Analogously to **2a**, compound **2f** (295 mg, 0.56 mmol, 89%) was obtained from **1** (160 mg, 0.63 mmol), Et_3N (100 μL, 0.72 mmol), 2,4,6-trichlorobenzoyl chloride (111 μL, 0.72 mmol), dihydrotestosterone (220 mg, 0.76 mmol), and DMAP (155 mg, 1.26 mmol) as a colourless solid of mp: 121 °C; R_f = 0.22 (ethyl acetate/*n*-hexane 1:2); ν_{max} (ATR): $\tilde{\nu}$ = 2934, 1713, 1596, 1517, 1387, 1293, 1284, 1170, 1126, 1024 cm^{-1}; ^1H NMR (300 MHz, $CDCl_3$): δ = 0.7–2.4 (37H, m, CMe_3, steroid-H), 4.44 (2H, d, ^3J = 5.6 Hz, CH_2N), 4.80 (1H, t, ^3J = 8.3 Hz, 17-H), 5.5–5.7 (1H, m, NH), 7.31 (1H, d, ^3J = 8.1 Hz, 5′-H), 8.19 (1H, d, ^3J = 8.1 Hz, 4′-H), 9.08 ppm (1H, s, 2′-H); ^{13}C NMR (75.5 MHz, $CDCl_3$): δ = 11.4 (13-Me), 12.3 (10-Me), 20.8 (C-11), 23.6 (C-15), 27.6 (C-6), 28.3 (CMe_3), 28.7 (C-16), 31.1 (C-7), 35.1 (C-8), 35.6 (C-10), 36.9 (C-12), 38.0 (C-2), 38.4 (C-1), 43.0 (C-13), 44.5 (C-4), 45.7 (CH_2N), 46.5 (C-5), 50.5 (C-9), 53.6 (C-14), 79.6 (CMe_3), 83.6 (C-17), 121.0 (C-5′), 125.1 (C-3′), 137.6 (C-4′), 150.3 (C-2′), 155.9 (OCON), 161.9 (C-6′), 165.0 (CO_2), 211.7 ppm (CO); *m/z* (EI) 469 (20), 450 (35), 178 (72), 161 (38), 135 (100).

*6′-(tert-Butoxycarbonylaminomethyl)nicotinic acid 3-oxoandrost-4-en-17β-yl ester (**2g**):* Analogously to **2a**, compound **2g** (282 mg, 0.56 mmol, 86%) was obtained from **1** (160 mg, 0.63 mmol), Et_3N (100 μL, 0.72 mmol), 2,4,6-trichlorobenzoyl chloride (111 μL, 0.72 mmol), testosterone (220 mg, 0.76 mmol), and DMAP (155 mg, 1.26 mmol) as a colourless solid of mp: 95 °C; R_f = 0.17 (ethyl acetate/*n*-hexane 1:2); ν_{max} (ATR): $\tilde{\nu}$ = 2937, 1713, 1670, 1598, 1274, 1167, 1118, 1023, 867, 759 cm^{-1}; ^1H NMR (300 MHz, $CDCl_3$): δ = 0.9–2.5 (34H, m, CMe_3, steroid-H), 4.47 (2H, d, ^3J = 5.6 Hz, CH_2N), 4.83 (1H, t, ^3J = 8.4 Hz, 17-H), 5.5–5.6 (1H, m, NH), 5.71 (1H, s, 4-H), 7.33 (1H, d, ^3J = 8.1 Hz, 5′-H), 8.21 (1H, d, ^3J = 8.1 Hz, 4′-H), 9.10 ppm (1H, s, 2′-H); ^{13}C NMR (75.5 MHz, $CDCl_3$): δ = 12.3 (13-Me), 17.4 (10-Me), 20.5 (C-11), 23.6 (C-15), 27.6 (C-16), 28.3 (CMe_3), 31.5 (C-7), 32.7 (C-6), 33.9 (C-2), 35.4 (C-8), 35.7 (C-1), 36.7 (C-12), 38.6 (C-10), 42.9 (C-13), 45.8 (CH_2N), 50.2 (C-14), 53.7 (C-9), 79.8 (CMe_3), 83.4 (C-17), 121.1 (C-5′), 124.0 (C-4), 125.1 (C-3′), 137.7 (C-4′), 150.4 (C-2′), 155.9 (OCON), 162.0 (C-6′), 165.0 (CO_2), 170.8 (C-5), 199.4 ppm (CO); *m/z* (EI) 467 (53), 448 (12), 423 (14), 179 (47), 151 (40), 135 (100).

2. Syntheses of 6-aminomethylnicotinates (3): *6′-Aminomethylnicotinic acid 17-oxoestra-1,3,5(10)-trien-3-yl ester dihydrochloride (**3a**):* Compound **2a** (270 mg, 0.54 mmol) was treated with 4 M HCl/dioxane (20 mL) at room temperature for 1 h. The precipitate formed was collected, washed with dioxane and diethyl ether, and dried. Yield: 218 mg (0.46 mmol, 85%); mp: 200 °C (dec.); ν_{max} (ATR): $\tilde{\nu}$ = 3043, 2931, 2856, 1733, 1645, 1491, 1272, 1209, 1082, 885, 715 cm^{-1}; ^1H NMR (300 MHz, $[D_6]DMSO/D_2O$): δ = 0.75 (3H, s, Me), 1.2–2.5 (16H, m), 2.7–2.8 (2H, m, 6-H), 4.3–4.4 (2H, m, CH_2N), 6.8–7.0 (2H, m, 2-H, 4-H), 7.27 (1H, d, ^3J = 8.4 Hz, 1-H), 7.52 (1H, d, ^3J = 8.2 Hz, 5′-H), 8.39 (1H, d, ^3J = 8.2 Hz, 4′-H), 9.14 ppm (1H, s, 2′-H); *m/z* (EI) 404 (10) [M^+ − 2HCl], 270 (69), 185 (27), 146 (23), 135 (100); accurate mass (EIMS) for $C_{25}H_{28}N_2O_3$ (free base): calcd 404.20999, obsd 404.21000.

*6′-Aminomethylnicotinic acid 17β-hydroxyestra-1,3,5(10)-trien-3-yl ester dihydrochloride (**3b**):* 176 mg (0.37 mmol, 79%) from **2b′** (290 mg, 0.47 mmol), analogously to the synthesis of **3a**; colourless solid of mp: 273 °C; ν_{max} (ATR): $\tilde{\nu}$ = 3295, 2925, 2860, 1741, 1645, 1492, 1287, 1082 cm^{-1}; ^1H NMR (300 MHz, $[D_6]DMSO$): δ = 0.68 (3H, s, 13-Me), 1.1–2.4 (13H, m, steroid-H), 2.8–2.9 (2H, m, 6-H), 3.53 (1H, t, ^3J = 8.5 Hz, 17-H), 4.3–4.4 (2H, m, CH_2N), 6.99 (1H, s, 4-H), 7.05 (1H, d, ^3J = 8.4 Hz, 2-H), 7.36 (1H, d, ^3J = 8.4 Hz, 1-H), 7.75 (1H, d, ^3J = 8.2 Hz, 5′-H), 8.50 (1H, d, ^3J = 8.2 Hz, 4′-H), 8.6–8.8 (3H, m, NH_3), 9.22 ppm (1H, s, 2′-H); ^{13}C NMR (75.5 MHz, $[D_6]DMSO$): δ = 11.3 (13-Me), 22.8 (C-15), 25.9 (C-11), 26.6 (C-7), 29.0 (C-6), 29.9 (C-16), 36.6 (C-12), 38.2 (C-8), 42.7 (C-13), 42.8 (CH_2N), 43.7 (C-9), 49.6 (C-14), 80.0 (C-17), 118.8 (C-2), 121.5 (C-4), 122.8 (C-5′), 124.8 (C-3′), 126.4 (C-1), 138.0 (C-4′), 138.2 (C-5), 138.3 (C-10), 148.0 (C-3), 149.8 (C-2′), 158.4 (C-6′), 163.5 ppm (CO_2); *m/z* (EI) 406 (45) [M^+ − 2HCl], 271 (21), 135 (100), 93 (52); accurate mass (EIMS) for $C_{25}H_{30}N_2O_3$ (free base): calcd 406.22564, obsd 406.22560.

*6′-Aminomethylnicotinic acid 20-oxopregna-5-en-3β-yl ester dihydrochloride (**3c**):* A solution of **1** (160 mg, 0.63 mmol) in DMF (2 mL) was treated with Et_3N (100 μL, 0.72 mmol) and 2,4,6-trichlorobenzoyl chloride (111 μL, 0.72 mmol). The resulting suspension was stirred at room temperature for 20 min. Pregnenolone (402 mg, 1.26 mmol) and DMAP (155 mg, 1.26 mmol) were added as a solution in toluene (20 mL), and the reaction mixture was stirred at room temperature for 16 h. After dilution with ethyl acetate and washing with water, the organic phase was dried over Na_2SO_4 and concentrated. After purification by column chromatography, (silica gel 60; ethyl acetate/*n*-hexane 1:2) the product was dissolved in THF (5 mL), 4 M HCl/dioxane (25 mL) was added, and the mixture was stirred at room temperature for 1 h. The resulting dihydrochloride salt was collected, washed with dioxane and diethyl ether, and dried. Yield: 182 mg (0.35 mmol, 55%); mp: 150 °C (dec.); ν_{max} (ATR): $\tilde{\nu}$ = 2941, 1726, 1703, 1644, 1356, 1293, 1120, 872, 754 cm^{-1}; ^1H NMR (300 MHz, $[D_6]DMSO/D_2O$): δ = 0.43 (3H, s, COMe), 0.7–2.4 (26H, m), 4.2–4.3 (2H, m, CH_2N), 4.57 (1H, m, 3-H), 5.32 (1H, m, 6-H), 7.46 (1H, d, ^3J = 8.0 Hz, 5′-H), 8.25 (1H, d, ^3J = 8.0 Hz, 4′-H), 8.99 ppm (1H, s, 2′-H); *m/z* (EI) 408 (2), 298 (18), 283 (4), 44 (100).

*6′-Aminomethylnicotinic acid 17-oxoandrost-5-en-3β-yl ester dihydrochloride (**3d**):* 277 mg (0.56 mmol, 98%) from **2d** (300 mg, 0.57 mmol), analogously to the synthesis of **3a**; colourless solid of mp: 190 °C (dec.); ν_{max} (ATR): $\tilde{\nu}$ = 3051, 2945, 1728, 1644, 1373, 1289, 1129 cm^{-1}; ^1H NMR (300 MHz, $[D_6]DMSO/D_2O$): δ = 0.78 (3H, s, 13-Me), 0.9–2.5 (22H, m), 4.2–4.3 (2H, m, CH_2N), 4.6–4.8 (1H, m, 3-H), 5.3–5.5 (1H, m, 6-H), 7.59 (1H, d, ^3J = 8.2 Hz, 5′-H), 8.31 (1H, d, ^3J = 8.2 Hz, 4′-H), 9.06 ppm (1H, s, 2′-H); ^{13}C NMR (75.5 MHz, $[D_6]DMSO/D_2O$): δ = 13.7 (13-Me), 19.6 (10-Me), 20.4 (C-11), 22.0 (C-

15), 27.8 (C-2), 30.8 (C-12), 31.5 (C-7), 31.6 (C-8), 36.0 (C-16), 36.8 (C-10), 36.9 (C-4), 38.0 (C-1), 42.9 (CH_2N), 47.6 (C-13), 50.1 (C-9), 51.3 (C-14), 75.4 (C-3), 122.7 (C-5′), 123.1 (C-6), 126.1 (C-3′), 138.4 (C-4′), 140.0 (C-5), 150.0 (C-2′), 157.7 (C-6′), 164.6 (CO_2), 221.7 ppm (CO); *m/z* (EI) 404 (76), 270 (50), 136 (41), 91 (100).

*6′-Aminomethylnicotinic acid 3-(tert-butyldimethylsilyloxy)estra-1,3,5(10)-trien-17β-yl ester dihydrochloride (**3e′**):* 232 mg (0.39 mmol, 78%) from **2e** (310 mg, 0.50 mmol), analogously to the synthesis of **3a**; colourless solid of mp: 237 °C; ν_{max} (ATR): $\tilde{\nu}$ = 2929, 2860, 1730, 1606, 1497, 1298, 1130, 953, 839, 779 cm^{-1}; ^1H NMR (300 MHz, [D_6]DMSO): δ = 0.13 (6H, s, SiMe), 0.9–1.0 (12H, m, $SiCMe_3$, 13-Me), 1.2–2.4 (13H, m, steroid-H), 2.7–2.8 (2H, m, 6-H), 4.2–4.3 (2H, m, CH_2N), 4.87 (1H, t, ^3J = 8.4 Hz, 17-H), 6.51 (1H, s, 4-H), 6.57 (1H, d, ^3J = 8.5 Hz, 2-H), 7.12 (1H, d, ^3J = 8.5 Hz, 1-H), 7.62 (1H, d, ^3J = 8.2 Hz, 5′-H), 8.35 (1H, d, ^3J = 8.2 Hz, 4′-H), 9.08 ppm (1H, s, 2′-H); ^{13}C NMR (75.5 MHz, [D_6]DMSO): δ = −3.0 (SiMe), 12.4 (13-Me), 18.0 ($SiCMe_3$), 23.2 (C-15), 26.1 ($SiCMe_3$), 27.1 (C-7), 27.6 (C-16), 29.4 (C-6), 36.8 (C-12), 42.7 (CH_2N), 43.3 (C-13), 43.5 (C-9), 49.3 (C-14), 83.8 (C-17), 113.0 (C-2), 115.2 (C-4), 122.9 (C-5′), 125.7 (C-3′), 130.6 (C-10), 137.5 (C-5), 138.1 (C-4′), 149.6 (C-2′), 155.0 (C-3), 157.7 (C-6′), 164.7 ppm (CO_2); *m/z* (EI) 520 (86), 463 (100), 209 (44); accurate mass (EIMS) for $C_{31}H_{44}N_2O_3Si$ (free base): calcd 520.31212, obsd 520.31210.

*6′-Aminomethylnicotinic acid 3-hydroxyestra-1,3,5(10)-trien-17β-yl ester dihydrotrifluoroacetate (**3e**):* A solution of **3e′** (75 mg, 0.13 mmol) in TFA/CH_2Cl_2 (1:1, 20 mL) was stirred at room temperature for 8 h. The solvent was evaporated, and the oily residue was dissolved in a small amount of dioxane. Diethyl ether/*n*-hexane was added, and the resulting colourless precipitate was collected, washed with diethyl ether, and dried. Yield: 50 mg (0.07 mmol, 54%); colourless solid of mp: 145 °C; ν_{max} (ATR): $\tilde{\nu}$ = 3266, 2918, 1729, 1683, 1603, 1498, 1365, 1289, 1202, 1184, 1125 cm^{-1}; ^1H NMR (300 MHz, [D_6]DMSO): δ = 0.93 (3H, s, 13-Me), 1.2–2.3 (13H, m, steroid-H), 2.7–2.8 (2H, m, 6-H), 4.3–4.4 (2H, m, CH_2N), 4.89 (1H, t, ^3J = 8.4 Hz, 17-H), 6.45 (1H, s, 4-H), 6.51 (1H, d, ^3J = 8.5 Hz, 2-H), 7.05 (1H, d, ^3J = 8.5 Hz, 1-H), 7.65 (1H, d, ^3J = 8.2 Hz, 5′-H), 8.2–8.3 (3H, m, NH_3), 8.38 (1H, d, ^3J = 8.2 Hz, 4′-H), 9.02 (1H, s, OH), 9.10 ppm (1H, s, 2′-H); ^{13}C NMR (75.5 MHz, [D_6]DMSO): δ = 11.2 (13-Me), 22.9 (C-15), 25.9 (C-11), 26.8 (C-7), 27.3 (C-16), 29.1 (C-6), 36.4 (C-12), 38.3 (C-8), 42.6 (CH_2N), 42.9 (C-13), 43.2 (C-9), 49.0 (C-14), 83.4 (C-17), 112.8 (C-2), 115.0 (C-4), 122.6 (C-5′), 125.4 (C-3′), 126.1 (C-1), 130.1 (C-10), 137.1 (C-5), 137.8 (C-4′), 149.3 (C-2′), 155.0 (C-6′), 155.0 (C-3), 157.8 (C-6′), 164.3 ppm (CO_2); *m/z* (EI) 406 (75) [$M^+ - 2F_3CCO_2H$], 153 (35), 135 (100); accurate mass (EIMS) for $C_{25}H_{30}N_2O_3$ (free base): calcd 406.22564, obsd 406.22560.

*6′-Aminomethylnicotinic acid 3-oxoandrostan-17β-yl ester dihydrochloride (**3f**):* 235 mg (0.47 mmol, 86%) from **2f** (290 mg, 0.55 mmol), analogously to the synthesis of **3a**; colourless solid of mp: 210 °C (dec.); ν_{max} (ATR): $\tilde{\nu}$ = 2918, 2849, 1729, 1719, 1646, 1447, 1357, 1304, 1298, 1128, 1004, 890, 758 cm^{-1}; ^1H NMR (300 MHz, [D_6]DMSO): δ = 0.7–2.5 (28H, m, steroid-H), 4.2–4.3 (2H, m, CH_2N), 4.79 (1H, t, ^3J = 8.3 Hz, 17-H), 7.69 (1H, d, ^3J = 8.2 Hz, 5′-H), 8.34 (1H, d, ^3J = 8.2 Hz, 4′-H), 8.5–8.8 (3H, m, NH_3), 9.07 ppm (1H, s, 2′-H); ^{13}C NMR (75.5 MHz, [D_6]DMSO): δ = 11.1 (13-Me), 12.2 (10-Me), 20.5 (C-11), 23.2 (C-15), 27.3 (C-6), 28.2 (C-16), 30.8 (C-7), 34.7 (C-10), 35.3 (C-8), 36.4 (C-12), 37.7 (C-2), 37.9 (C-1), 42.7 (C-4, C-13), 44.2 (CH_2N), 46.0 (C-5), 49.9 (C-9), 53.0 (C-14), 83.4 (C-17), 122.7 (C-5′), 125.4 (C-3′), 137.7 (C-4′), 149.2 (C-2′), 157.9 (C-6′), 164.3 (CO_2), 210.4 ppm (CO); *m/z* (EI) 424 (4) [$M^+ - 2HCl$], 135 (20), 36 (100); accurate mass (EIMS) for $C_{26}H_{36}N_2O_3$ (free base): calcd 424.27259, obsd 424.27260.

*6′-Aminomethylnicotinic acid 3-oxo-androst-4-en-17β-yl ester dihydrochloride (**3g**):* 212 mg (0.47 mmol, 86%) from **2g** (263 mg, 0.50 mmol), analogously to the synthesis of **3a**; colourless solid of mp: 190 °C (dec.); ν_{max} (ATR): $\tilde{\nu}$ = 2940, 2849, 1730, 1715, 1684, 1645, 1479, 1357, 1331, 1306, 1295, 1126, 1086, 998, 890, 758 cm^{-1}; ^1H NMR (300 MHz, [D_6]DMSO): δ = 0.9–2.5 (25H, m, steroid-H), 4.2–4.4 (2H, m, CH_2N), 4.80 (1H, t, ^3J = 8.3 Hz, 17-H), 5.64 (1H, s, 4-H), 7.70 (1H, d, ^3J = 8.2 Hz, 5′-H), 8.34 (1H, d, ^3J = 8.2 Hz, 4′-H), 8.6–8.8 (3H, m, NH_3), 9.07 ppm (1H, s, 2′-H); ^{13}C NMR (75.5 MHz, [D_6]DMSO): δ = 12.1 (13-Me), 16.9 (10-Me), 20.1 (C-11), 23.2 (C-15), 27.2 (C-16), 31.2 (C-7), 31.9 (C-6), 33.6 (C-2), 34.7 (C-8), 35.1 (C-1), 36.2 (C-12), 38.2 (C-10), 42.5 (C-13), 42.7 (CH_2N), 49.4 (C-14), 53.0 (C-9), 83.2 (C-17), 122.7 (C-5′), 123.2 (C-4), 125.3 (C-3′), 137.7 (C-4′), 149.2 (C-2′), 157.9 (C-6′), 164.3 (CO_2), 170.8 (C-5), 198.0 ppm (CO); *m/z* (EI) 422 (18) [$M^+ - 2HCl$], 406 (41), 135 (57), 120 (100), 92 (48); accurate mass (EIMS) for $C_{26}H_{34}N_2O_3$ (free base): calcd 422.25694, obsd 422.25703.

3. Syntheses of the platinum complexes (4): *cis-Dichloro(6′-aminomethylnicotinic 17-oxoestra-1,3,5(10)-trien-3-yl ester)platinum(II) (**4a**):* Compound **3a** (196 mg, 0.41 mmol) was dissolved in H_2O/THF (10 mL, 1:1). K_2PtCl_4 (170 mg, 0.41 mmol), dissolved in H_2O, was added, and the colourless precipitate was redissolved by addition of THF. The pH value was adjusted to 5–6 with aqueous NaOH, and the reaction mixture was stirred at room temperature for 24 h. The yellow precipitate was collected, washed in turn with water, acetone, and diethyl ether, and finally dried. Yield: 210 mg (0.31 mmol, 76%); yellow solid of mp: >250 °C; $C_{25}H_{28}Cl_2N_2O_3Pt$ requires: C 44.79, H 4.21, N 4.18; found: C 44.53, H 4.11, N 4.03; ν_{max} (ATR): $\tilde{\nu}$ = 3210, 2927, 1738, 1622, 1580, 1492, 1270, 1256, 1155, 1083, 742 cm^{-1}; ^1H NMR (300 MHz, [D_7]DMF): δ = 0.92 (3H, s, 13-Me), 1.4–2.6 (13H, m), 2.9–3.0 (2H, m, 6-H, hidden), 4.53 (2H, t, ^3J = 5.9 Hz, CH_2N), 6.3–6.5 (2H, m, NH_2), 7.0–7.2 (2H, m, 2-H, 4-H), 7.44 (1H, d, ^3J = 8.4 Hz, 1-H), 7.99 (1H, d, ^3J = 8.3 Hz, 5′-H), 8.83 (1H, d, ^3J = 8.3 Hz, 4′-H), 10.0 ppm (1H, s, 2′-H); ^{13}C NMR (75.5 MHz, [D_7]DMF): δ = 13.7 (13-Me), 21.6 (C-15), 26.0 (C-11), 26.4 (C-7), 29.5 (C-6), 32.0 (C-12), 35.7 (C-16), 38.3 (C-8), 44.4 (C-9), 47.9 (CH_2N), 50.4 (C-14), 53.8 (C-13), 119.2 (C-2), 121.8 (C-4), 122.7 (C-5′), 126.8 (C-3′), 126.9 (C-1), 138.5 (C-5), 138.7 (C-10), 139.3 (C-4′), 148.8 (C-2′), 148.9 (C-3), 162.5 (C-6′), 171.3 (CO_2), 219.6 ppm (CO); ^{195}Pt NMR (64.4 MHz, [D_7]DMF): δ = 2441 ppm; *m/z* (EI) 400 (3), 270 (10), 172 (8), 131 (16), 106 (40), 36 (100).

*cis-Dichloro(6′-aminomethylnicotinic 17-hydroxyestra-1,3,5(10)-trien-3-yl ester)platinum(II) (**4b**) from **2b**:* A solution of **2b** (56 mg, 0.11 mmol) in CH_2Cl_2 (5 mL) was treated with trifluoroacetic acid (5 mL), and the resulting mixture was stirred at room temperature for 1 h. The solvent was evaporated, and the residue was dissolved in H_2O/THF (5 mL, 3:2). K_2PtCl_4 (46 mg, 0.11 mmol), dissolved in water, was added, and the colourless precipitate was redissolved by addition of THF. After stirring at room temperature for 24 h the yellow solid was collected, washed with water, acetone, and diethyl ether, and dried. Yield: 32 mg (0.05 mmol, 43%); mp: >250 °C; $C_{25}H_{30}Cl_2N_2O_3Pt$ requires: C 44.65, H 4.50, N 4.17; found: C 44.66, H 4.99, N 4.05; ν_{max} (ATR): $\tilde{\nu}$ = 3464, 3205, 2920, 2870, 1743, 1620, 1493, 1269, 1213, 1154, 1053, 744 cm^{-1}; ^1H NMR (300 MHz, [D_7]DMF): δ = 0.78 (3H, s, 13-Me), 1.2–2.5 (13H, m), 2.8–2.9 (2H, m, 6-H, hidden), 3.6–3.7 (1H, m, 17-H), 4.53 (2H, t, ^3J = 5.9 Hz, CH_2N), 4.59 (1H, d, ^3J = 4.8 Hz, 17-OH), 6.3–6.5 (2H, m, NH_2), 7.0–7.2 (2H, m, 2-H, 4-H), 7.43 (1H, d, ^3J = 8.5 Hz, 1-H), 7.98 (1H, d, ^3J = 8.3 Hz, 5′-H), 8.83 (1H, d, ^3J = 8.3 Hz, 4′-H), 9.99 ppm (1H, s, 2′-H); ^{13}C NMR (75.5 MHz, [D_7]DMF): δ = 11.2 (13-Me), 23.3 (C-15), 26.5 (C-11), 27.2 (C-7), 30.4 (C-6), 30.6 (C-16), 37.2 (C-12), 38.9 (C-8), 43.4 (C-13), 44.5 (C-9), 50.4 (C-14), 53.8 (CH_2N), 81.0 (C-17), 119.0 (C-2), 121.8 (C-4),

122.7 (C-5′), 126.8 (C-3′), 126.8 (C-1), 138.7 (C-5), 139.0 (C-10), 139.3 (C-4′), 148.7 (C-3), 148.8 (C-2′), 162.5 (C-6′), 171.3 ppm (CO_2); ^{195}Pt NMR (64.4 MHz, [D$_7$]DMF): $\delta = 2440$ ppm; *m/z* (EI) 406 (12) [$M^+ - PtCl_2$], 402 (13), 368 (42), 272 (99), 213 (54), 172 (75), 106 (87), 91 (75), 36 (100).

*Complex **4b** from **3b**:* Analogously to **4a**, compound **4b** (123 mg, 0.18 mmol, 81%) was obtained from **3b** (106 mg, 0.22 mmol) and K_2PtCl_4 (92 mg, 0.22 mmol) as a yellow solid of mp: >250 °C; *m/z* (EI) 406 (12), 402 (13), 368 (42), 272 (99), 213 (54), 172 (75), 106 (87), 91 (75), 36 (100).

*cis-Dichloro(6′-aminomethylnicotinic 20-oxopregna-5-en-3β-yl ester)-platinum(II) (**4c**):* Analogously to **4a**, compound **4c** (110 mg, 0.15 mmol, 62%) was obtained from **3c** (131 mg, 0.25 mmol) and K_2PtCl_4 (104 mg, 0.25 mmol) as a yellow solid of mp: >250 °C; $C_{28}H_{38}Cl_2N_2O_3Pt$ requires: C, 46.93, H 5.34, N 3.91; found: C 46.84, H 5.24, N 4.03; ν_{max} (ATR): $\tilde{\nu} = 3232$, 2936, 1726, 1706, 1622, 1575, 1293, 1275, 1131 cm^{-1}; ^{1}H NMR (300 MHz, [D$_7$]DMF): $\delta = 0.62$ (3H, s, COMe), 0.9–2.2 (26H, m), 2.51 (2H, d, $^3J = 7.4$ Hz, 4-H), 2.6–2.7 (1H, m, 17-H), 4.48 (2H, t, $^3J = 5.6$ Hz, CH_2N), 4.7–4.9 (1H, m, 3-H), 5.4–5.5 (1H, m, 6-H), 6.3–6.4 (2H, m, NH_2), 7.91 (1H, d, $^3J = 8.3$ Hz, 5′-H), 8.68 (1H, d, $^3J = 8.3$ Hz, 4′-H), 9.85 ppm (1H, s, $^3J_{PtH}$ 33 Hz, 2′-H); ^{13}C NMR (75.5 MHz, [D$_7$]DMF): $\delta = 13.0$ (13-Me), 19.2 (10-Me), 21.2 (C-11), 22.8 (C-16), 24.5 (C-15), 27.8 (C-2), 31.1 (C-8), 31.9 (C-7), 32.0 (CO*Me*), 36.8 (C-10), 37.1 (C-1), 38.0 (C-4), 38.7 (C-12), 43.8 (C-13), 50.2 (C-9), 53.7 (CH_2N), 56.8 (C-14), 63.3 (C-17), 76.1 (C-3), 122.5 (C-5′), 122.9 (C-6), 127.4 (C-3′), 138.9 (C-4′), 139.9 (C-5), 148.5 (C-2′), 162.9 (C-6′), 170.8 (CO_2), 208.5 ppm (CO); ^{195}Pt NMR (64.4 MHz, [D$_7$]DMF): $\delta = 2440$ ppm; *m/z* (EI) 298 (8) [$C_{21}H_{30}O^+$], 283 (2), 91 (46), 43 (100).

*cis-Dichloro(6′-aminomethylnicotinic acid 17-oxoandrost-5-en-3β-yl ester)platinum(II) (**4d**):* Analogously to **4a**, compound **4d** (179 mg, 0.26 mmol, 87%) was obtained from **3d** (150 mg, 0.30 mmol) and K_2PtCl_4 (125 mg, 0.30 mmol) as a yellow solid of mp: >250 °C; $C_{26}H_{34}Cl_2N_2O_3Pt$ requires: C 45.35, H 4.98, N 4.07; found: C 44.93, H 5.05, N 3.92; ν_{max} (ATR): $\tilde{\nu} = 3236$, 2946, 1736, 1724, 1622, 1406, 1293, 1277, 1134, 751 cm^{-1}; ^{1}H NMR (300 MHz, [D$_7$]DMF): $\delta = 0.88$ (3H, s, 13-Me), 1.0–2.6 (22H, m), 4.49 (2H, t, $^3J = 5.9$ Hz, CH_2N), 4.7–4.9 (1H, m, 3-H), 5.4–5.6 (1H, m, 6-H), 6.3–6.4 (2H, m, NH_2), 7.91 (1H, d, $^3J = 8.2$ Hz, 5′-H), 8.67 (1H, d, $^3J = 8.2$ Hz, 4′-H), 9.83 ppm (1H, s, 2′-H); ^{13}C NMR (75.5 MHz, [D$_7$]DMF): $\delta = 13.3$ (13-Me), 19.2 (10-Me), 20.5 (C-11), 21.9 (C-15), 27.8 (C-2), 30.8 (C-12), 31.6 (C-7), 31.8 (C-8), 35.6 (C-16), 36.9 (C-4), 37.0 (C-10), 38.0 (C-1), 47.4 (C-13), 50.5 (C-9), 51.7 (C-14), 53.7 (CH_2N), 76.0 (C-3), 122.5 (C-5′), 122.6 (C-6), 127.3 (C-3′), 138.9 (C-4′), 140.1 (C-5), 148.4 (C-2′), 162.6 (C-6′), 170.8 (CO_2), 219.7 ppm (CO); ^{195}Pt NMR (64.4 MHz, [D$_7$]DMF): $\delta =$ 2440 ppm; *m/z* (EI) 270 (62), 91 (66), 79 (44), 36 (100).

*cis-Dichloro(6′-aminomethylnicotinic acid 3-hydroxyestra-1,3,5(10)-trien-17β-yl ester) platinum(II) (**4e**):* Analogously to **4a**, compound **4e** (30 mg, 0.045 mmol, 69%) was obtained from **3e** (41 mg, 0.065 mmol) and K_2PtCl_4 (27 mg, 0.065 mmol) as a yellow solid of mp: >250 °C; $C_{25}H_{30}Cl_2N_2O_3Pt$ requires: C 44.65, H 4.50, N 4.17; found: C 44.66, H 4.69, N 3.95; ν_{max} (ATR): $\tilde{\nu} = 3189$, 2923, 2862, 1711, 1607, 1498, 1292, 1126 cm^{-1}; ^{1}H NMR (300 MHz, [D$_7$]DMF): $\delta = 0.98$ (3H, s, 13-Me), 1.3–2.4 (13H, m, steroid-H), 2.7–2.8 (2H, m, 6-H, hidden), 4.48 (2H, t, $^3J = 5.9$ Hz, CH_2N), 4.92 (1H, t, $^3J = 8.3$ Hz, 17-H), 6.3–6.5 (2H, m, NH_2), 6.56 (1H, s, 4-H), 6.63 (1H, d, $^3J =$ 8.4 Hz, 2-H), 7.11 (1H, d, $^3J = 8.4$ Hz, 1-H), 7.92 (1H, d, $^3J = 8.2$ Hz, 5′-H), 8.69 (1H, d, $^3J = 8.2$ Hz, 4′-H), 9.19 (1H, s, OH), 9.93 ppm (1H, s, 2′-H); ^{13}C NMR (75.5 MHz, [D$_7$]DMF): $\delta = 12.2$ (13-Me), 23.4 (C-15), 26.5 (C-11), 27.5 (C-7), 27.8 (C-16), 30.4 (C-6), 37.1 (C-12), 39.0 (C-8), 43.5 (C-13), 44.0 (C-9), 49.7 (C-14), 53.7 (CH_2N), 84.7 (C-17), 113.2 (C-2), 115.4 (C-4), 121.5 (C-5′), 126.5 (C-1), 127.3 (C-3′), 130.8 (C-10), 137.3 (C-5), 137.8 (C-4′), 148.5 (C-2′), 155.9 (C-3), 163.4 (C-6′), 170.7 ppm (CO_2); ^{195}Pt NMR (64.4 MHz, [D$_7$]DMF): $\delta = 2439$ ppm; *m/z* (EI) 254 (62), 159 (40), 146 (45), 133 (56), 36 (100).

*cis-Dichloro(6′-aminomethylnicotinic acid 3-oxoandrostan-17β-yl ester)platinum(II) (**4f**):* Analogously to **4a**, compound **4f** (240 mg, 0.35 mmol, 87%) was obtained from **3f** (200 mg, 0.40 mmol) and K_2PtCl_4 (167 mg, 0.40 mmol) as a yellow solid of mp: >250 °C; $C_{26}H_{36}Cl_2N_2O_3Pt$ requires: C 45.22, H 5.25, N 4.06; found: C 44.91, H 5.45, N 3.93; ν_{max} (ATR): $\tilde{\nu} = 3193$, 2933, 1717, 1617, 1297, 1129, 753 cm^{-1}; ^{1}H NMR (300 MHz, [D$_7$]DMF): $\delta = 0.8$–2.5 (28H, m, steroid-H), 4.48 (2H, t, $^3J = 5.6$ Hz, CH_2N), 4.83 (1H, t, $^3J = 8.2$ Hz, 17-H), 6.3–6.4 (2H, m, NH_2), 7.91 (1H, d, $^3J = 8.2$ Hz, 5′-H), 8.67 (1H, d, $^3J =$ 8.2 Hz, 4′-H), 9.92 ppm (1H, s, 2′-H); ^{13}C NMR (75.5 MHz, [D$_7$]DMF): $\delta = 11.1$ (13-Me), 12.3 (10-Me), 21.1 (C-11), 23.7 (C-15), 27.8 (C-6), 28.9 (C-16), 31.4 (C-7), 35.4 (C-10), 35.9 (C-8), 37.0 (C-12), 38.1 (C-2), 38.7 (C-1), 43.3 (C-13), 44.6 (C-4), 46.8 (C-5), 50.6 (C-9), 53.7 (CH2N), 53.9 C-14), 84.7 (C-17), 122.6 (C-5′), 127.3 (C-3′), 138.8 (C-4′), 148.5 (C-2′), 163.3 (C-6′), 170.8 (CO2), 210.3 ppm (CO); ^{195}Pt NMR (64.4 MHz, [D$_7$]DMF): $\delta = 2440$ ppm; *m/z* (EI) 424 (8), 272 (39), 257 (38), 149 (34), 135 (28), 91 (39), 79 (42), 36 (100).

*cis-Dichloro(6′-aminomethylnicotinic acid 3-oxoandrost-4-en-17β-yl ester)platinum(II) (**4g**):* Analogously to **4a**, compound **4g** (175 mg, 0.26 mmol, 70%) was obtained from **3g** (184 mg, 0.37 mmol) and K_2PtCl_4 (154 mg, 0.37 mmol) as a yellow solid of mp: >250 °C; $C_{26}H_{36}Cl_2N_2O_3Pt$ requires: C 45.35, H 4.98, N 4.07; found: C 45.51, H 5.28, N 3.81; ν_{max} (ATR): $\tilde{\nu} = 3237$, 2937, 1729, 1671, 1617, 1297, 1128, 754 cm^{-1}; ^{1}H NMR (300 MHz, [D$_7$]DMF): $\delta = 0.9$–2.5 (25H, m, steroid-H), 4.48 (2H, t, $^3J = 6.0$ Hz, CH_2N), 4.83 (1H, t, $^3J = 8.4$ Hz, 17-H), 5.67 (1H, s, 4-H), 6.3–6.4 (2H, m, NH_2), 7.91 (1H, d, $^3J = 8.2$ Hz, 5′-H), 8.67 (1H, d, $^3J = 8.2$ Hz, 4′-H), 9.92 ppm (1H, s, 2′-H); ^{13}C NMR (75.5 MHz, [D$_7$]DMF): $\delta = 12.1$ (13-Me), 17.1 (10-Me), 20.7 (C-11), 23.6 (C-15), 27.7 (C-16), 31.8 (C-7), 32.5 (C-6), 34.1 (C-2), 35.4 (C-8), 35.9 (C-1), 36.8 (C-12), 38.8 (C-10), 43.1 (C-13), 50.2 (C-14), 53.7 (CH_2N), 54.0 (C-9), 84.5 (C-17), 122.6 (C-5′), 123.7 (C-4), 127.2 (C-3′), 138.8 (C-4′), 148.5 (C-2′), 163.3 (C-6′), 170.8 (CO_2), 171.1 (C-5), 198.2 ppm (CO); ^{195}Pt NMR (64.4 MHz, [D$_7$]DMF): $\delta = 2439$ ppm; *m/z* (EI) 422 (4), 135 (9), 36 (100).

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