and order the energies of conformations of molecules such as hydrocarbons and a variety of programs can execute this function. Furthermore, once the conformations have been ordered in terms of energies, one has confidence that the calculated lowest energy conformations are those which contribute to the ground-state properties of the molecule. This is due to the minimal charge and polarizability associated with hydrocarbons. The same cannot be said for molecules with more highly charged or polar atoms such as are found in peptides or proteins. As an example, suppose one generates conformations for a polypeptide and evaluates their energies in vacuo. It is highly probable that the most favored conformations will be those that maximize intramolecular electrostatic interactions and hydrogen bonding. If the energies of those same conformations were now evaluated in the presence of a polar solvent, a significant number of the previously favorable interactions would be replaced by more energetically favorable solvent–polypeptide interactions. The differences in the in vacuo and solvent calculations may be so notable that one would select totally different groups to represent low-energy conformations under the two conditions.

Thus, one would like to calculate the energies of conformations for some molecules in the presence of explicit solvent molecules. Such calculations, however, are often prohibitively expensive in terms of CPU time, even for molecular mechanical force fields. Approximations may be used to simulate the presence of solvent, but often these are also fairly crude and the ordering of the energies of conformations may be inaccurate. Although some recent work has been done in this area, for example in hydration shell calculations,[50] this problem obviously needs to be addressed further since it is foolish to spend a large amount of effort generating conformations if the subsequent energy evaluation is not representative of what is found in nature.

A great deal of work has been done and is currently being pursued in the field of conformational searching, of which we have presented only a sampling. For small and moderately sized systems, systematic searches based upon expert systems perhaps offer the most efficient and convenient approach. Larger molecules may be more amenable to distance geometry methods or molecular dynamics coupled with distance constraints derived from experiment. While these methods may not always exhaustively sample low-energy conformations in molecules, especially in the case of macromolecular systems, they do provide a means of generating conformations which can provide insight into understanding the physical properties of molecules.

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Articles

Platinum Complexes with Binding Affinity for the Estrogen Receptor

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A number of (1,2-diaminoethane)dichloroplatinum(II) complexes, linked to dihydroxy-2-phenylindole by spacer groups of varying lengths, were synthesized and studied for their binding affinities for the calf uterine estrogen receptor. Best binding conditions were provided by the *n*-hexyl and the *p*-xylene group as spacer with RBA values of 6.5 (**16c**) and 4.4 (**17c**), respectively (17β-estradiol: RBA = 100). These values are only slightly lower than those of the corresponding diaminoethane ligands.

Endocrine therapy of hormone-dependent mammary tumors has proven to be a valuable alternative to chemotherapy with cytostatic agents because of the low toxicity associated with drugs like antiestrogens or aromatase inhibitors.[1-3] However, this treatment is limited by the fact that approximately 40% of the patients with estrogen receptor positive tumors do not respond to endocrine manipulations.[4] The reason for this lack of response is still unclear. The presence of these receptors in tumors that do not respond initiated our search for new compounds that bind to the receptor but exert their antitumor effect by a different mode of action. Substances with receptor affinity and that carry a cytotoxic group were thought to be good candidates for this purpose. The diaminodichloroplatinum(II) group was chosen as the cytostatic function because the parent compound *cis*-platinum is a potent antineoplastic agent against some tumors, especially against testicular cancer, but with low activity against breast cancer. Receptor affinity of platinum complexes might make it possible to overcome the resistance of mammary tumors to *cis*-platinum. Our rationale is based solely on the presence of estrogen receptors in the malignant cells but not on their function as transmitters of hormonal signals. The receptor should only be used to direct the cytotoxic agent toward the target cell.

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Scheme I

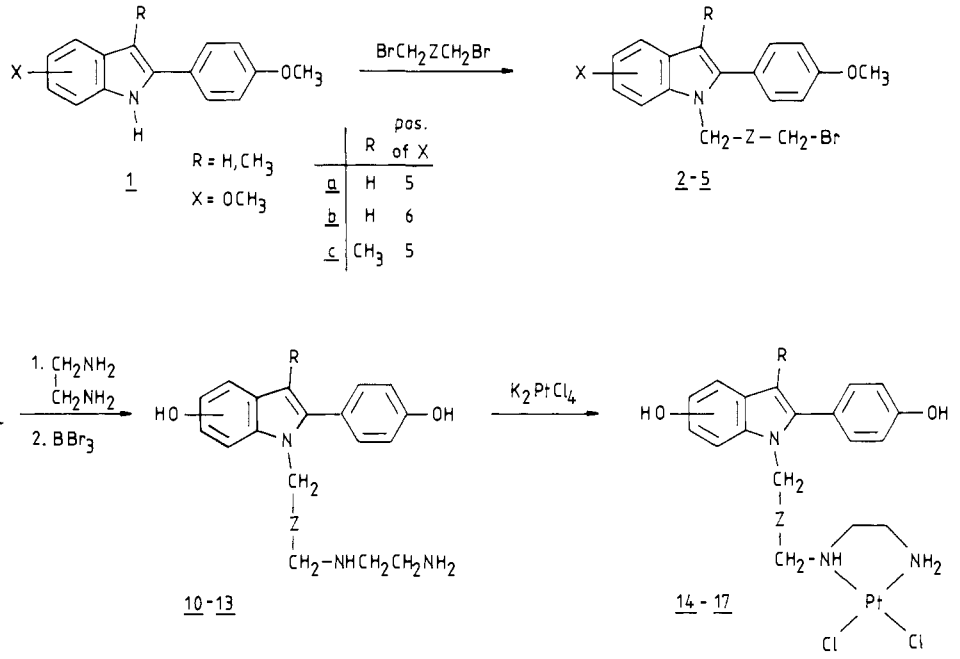
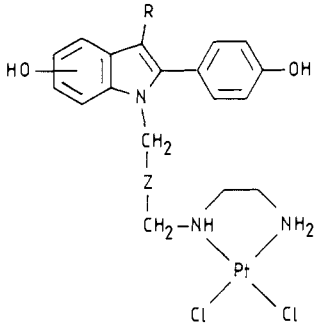


Chart I



From our previous work on the development of new antiestrogens[5-7] we knew that hydroxylated 2-phenylindoles are suitable structures for the synthesis of compounds binding to the estrogen receptor. The *cis*-(diaminoalkane)dichloroplatinum(II) group, which was applied as cytostatic moiety, was linked to the indole by a spacer group to avoid strong steric interference with the binding site. Since earlier studies[8,9] showed that the nitrogen is the best position for the introduction of bulky substituents into the indole, this atom was used to connect both moieties. As chelating function, the 1,2-diaminoethane group was applied with the spacer group fixed to one nitrogen (Chart I).

Chemistry. The starting dimethoxy-1*H*-2-phenylindoles **1a–d** were obtained by the Bischler indole synthesis as described previousy.[6] N-Substitution was accomplished by deprotonation with sodium hydride in DMF and subsequent addition of 1,ω-dibromoalkanes or α,α′-dibromo-1,4-xylene (Scheme I). Treatment of the (ω-bromoalkyl)indoles **2–5** with an excess of 1,2-diaminoethane yielded the 1-[[(2-aminoethyl)amino]alkyl]-2-phenylindoles **6–9**. Demethylation of the methoxy compounds was effected with BBr_3 in CH_2Cl_2. The crude products were purified by treatment with hot triethylamine solution. The diaminoderivatives **10–13** were converted into the dichloroplatinum(II) complexes **14–17** by the reaction with K_2PtCl_4 in aqueous DMF.

The structural assignment of all of the *cis*-diaminodichloroplatinum complexes was mainly made on the basis of their IR, NMR, and mass spectra. All of the complexes showed typical vibrations for Pt–N (435 cm^{-1}) and Pt–Cl (375, 330 sh). The most important feature in the ^1H NMR spectra is the occurrence of two separate multiplets together with two singlets for the phenolic hydroxy groups at low field. This pattern of signals can be explained by two different hydroxy groups and, additionally, by an interaction between these oxygens and the platinum atom and a long-range coupling with ring protons. Since these peaks are not influenced by the kind of spacer used, an intermolecular coordination has to be assumed. Temperature-dependent NMR spectroscopy showed that the two multiplets for the phenolic protons disappear at 120 °C whereas the singlets remain unchanged. At the same temperature, some of the splitting of the aromatic signals was lost. After cooling, the spectrum regains its original structure. These observations strongly support the assumption of an intermolecular association. Mass spectra of the complexes were obtained by application of the FAB method. The characteristic ions in the cation spectra are $[LPtCl_2]^+$, $[LPtCl]^+$, $[LPtCl\cdot DMSO]^+$, $[LPt]^+$, and $[LH]^+$ (L = ligand, DMSO derives from the matrix).

Binding Affinity for the Calf Uterine Estrogen Receptor. The binding affinities of both the diamino ligands (Table II) and the platinum complexes (Table III) were measured in a competitive binding assay with [^3H]-17β-estradiol. Calf uterine cytosol was used as receptor source and the dextran-coated charcoal (DCC) method was applied.[10] The relative binding affinities (RBA) are given as the ratio of the molar concentrations of 17β-estradiol and indole required to decrease the receptor-bound ra-

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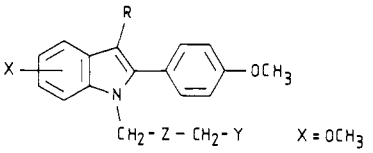
Table I. Dimethoxy-1-(bromoalkyl)- (Y = Br) and -1-[[(2-aminoethyl)amino]alkyl]-2-phenylindoles (Y = $NHCH_2CH_2NH_2$)



compd[a]	R	position of X	Z	formula[b]	mp,[c] °C	compd[d]	formula[e]
2a	H	5	$(CH_2)_2$	$C_{20}H_{22}BrNO_2$	74–76	**6a**	$C_{22}H_{29}N_3O_2$
2b	H	6	$(CH_2)_2$	$C_{20}H_{22}BrNO_2$	84–86	**6b**	$C_{22}H_{29}N_3O_2$
2c	CH_3	5	$(CH_2)_2$	$C_{21}H_{24}BrNO_2$	59–60	**6c**	$C_{23}H_{31}N_3O_2$
3c	CH_3	5	$(CH_2)_3$	$C_{22}H_{26}BrNO_2$	130–132	**7c**	$C_{24}H_{33}N_3O_2$
4a	H	5	$(CH_2)_4$	$C_{22}H_{26}BrNO_2$	82–83	**8a**	$C_{24}H_{33}N_3O_2$
4b	H	6	$(CH_2)_4$	$C_{22}H_{26}BrNO_2$	oil	**8b**	$C_{24}H_{33}N_3O_2$
4c	CH_3	5	$(CH_2)_4$	$C_{23}H_{28}BrNO_2$	60	**8c**	$C_{25}H_{35}N_3O_2$
5c	CH_3	5	C_6H_4	$C_{25}H_{24}BrNO_2$	109–111	**9c**	$C_{27}H_{31}N_3O_2$

[a] Y = Br. [b] Crystalline products were analyzed for C, H, and N within ±0.4% of the calculated values. [c] Recrystallized from EtOH. [d] Y = $NHCH_2CH_2NH_2$. [e] Compounds **6–9** are obtained as oils and purified by vacuum distillation.

dioactivity by 50%, multiplied by 100. The semilogarithmic plot of bound radioactivity vs molar concentrations of competitors exhibited curves parallel to those of 17β-estradiol, suggesting a common binding site for all of the compounds that were tested. All of the free diamino ligands are able to displace estradiol from its receptor. The RBA values range from 0.04 (**10b**) to 11.6 (**13c**). The methyl group in position 3 of the indole is essential for high binding affinity; therefore, usually only the 3-methylindole derivativs were synthesized. The RBA values increase considerably when the length of the alkyl spacer is varied from C_4 or C_5 to C_6. Derivatives with a short spacer group exhibit binding affinities similar to those of 2-phenylindoles carrying alkyl chains of the same length without amino functions.[6] The structural modification of shifting the hydroxy group from position 5 to C-6 of the indole decreases the receptor affinity and, therefore, was not generally performed. Contrary to our expectations, the binding affinities of these compounds were only slightly reduced following the conversion into the dichloroplatinum(II) complexes. The RBA values are in the same order of magnitude as other 2-phenylindoles that we have developed as antiestrogens.[6,7,11]

In this study, we have shown that it is possible to synthesize nonsteroidal (diaminoethane)dichloroplatinum(II) complexes with high binding affinity for the estrogen receptor that can potentially be used for the treatment of estrogen receptor containing tumors like mammary or prostatic carcinomas. We reached this goal by using a hydroxylated 2-phenylindole as binding moiety and a spacer group of appropriate length fixed to the indole nitrogen. The ethylenediamino group was used as chelating function for the platinum. The high affinities of the ligands **12c** and **13c** with a link of six carbon atoms between the amino groups and the indole nucleus make the interaction with an additional binding site off the receptor site likely. Complexes **16c** and **17c** exhibited binding affinities in the same order of magnitude as potent antiestrogens like tamoxifen (RBA = 2[12]) and the deacetylated zindoxifene (RBA = 9.5[7]). Interestingly, the binding affinities of the indole-derived complexes do not differ very much from those of the diamino ligands, whereas the ligand of the only steroidal platinum complex with high binding affinity for the estrogen receptor is devoid of affinity, probably, because it lacks the spacer group.[13] The endocrine properties and antitumor activity of the platinum complexes described in this paper will be published later.

Experimental Section

Melting points were determined on a Büchi 510 apparatus and are uncorrected. Elemental analyses were performed by the Mikroanalytisches Laboratorium, University of Regensburg. NMR spectra were obtained on a Varian EM 360L and a Bruker WM 250 spectrometer and are consistent with the assigned structures. Mass spectra were recorded on a Varian MAT CH 5 spectrometer. The syntheses of the starting indoles 5-methoxy-2-(4-methoxyphenyl) indole (1a), 6-methoxy-2-(4-methoxyphenyl)indole (1b), and 6-methoxy-2-(4-methoxyphenyl)-3-methylindole (1c) have been described previously.[6]

General Procedure for the Synthesis of 1-(Bromoalkyl)-2-phenylindoles 2–4 and 1-[4-(Bromomethyl)benzyl]-2-phenylindole (5). A solution of the 2-phenylindole (7.5 mmol) in 25 mL of dry DMF was added with stirring to a mixture of sodium hydride (12.5 mmol) in 40 mL of dry DMF at 0 °C. After stirring for 30 min at 0 °C, this mixture was added slowly to a solution of the dibromoalkane (11.0 mmol) in 25 mL of dry DMF with stirring and cooling. After the mixture was stirred for an additional 0.5 h, the excess of sodium hydride was destroyed by adding water. After extraction with CH_2Cl_2, the organic layer was dried with $MgSO_4$. After evaporation of the solvent in vacuo, the residiue was purified by column chromatography (SiO_2; CH_2Cl_2) and recrystallized from EtOH. The yields were in the range of 60–85%, except for **5** (25%). Melting points are reported in Table I.

General Procedure for the Synthesis of 1-[[(2-Aminoethyl)amino]alkyl]-2-phenylindoles 6–9. Under a nitrogen atmosphere, a solution of the (bromoalkyl)indole (4.0 mmol) in 80 mL of dry MeOH was added to 6.0 mmol of ethylenediamine in 40 mL of MeOH. After boiling for 12 h under reflux, water was added and the mixture extracted with CH_2Cl_2. The organic layer was dried with Na_2SO_4 and the solvent removed in vacuo. The resulting oil was purified by distillation in high vacuum. The yields were 70–80%.

General Procedure for the Ether Cleavage. A solution of the methoxy-substituted indole (4.0 mmol) in dry CH_2Cl_2 (100 mL) was cooled to -60 °C under a nitrogen atmosphere, and then BBr_3 (10.0 mmol) dissolved in 10 mL of CH_2Cl_2 was added. After removal of the cooling bath, the mixture was stirred over night, followed by boiling under reflux for 2 h. Under a nitrogen atmosphere and cooling with an ice bath, MeOH was added dropwise until the vigorous reaction ceased. After evaporation of the solvent, the residue was treated with saturated $NaHCO_3$ solution. The crude product was removed by filtration and extracted for 5 h with 200 mL of NEt_3 in a Soxhlet apparatus. The hot solution was filtered and the solvent removed in vacuo. The pure product was obtained by resuspension in water and filtration. The yields

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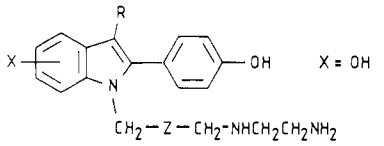
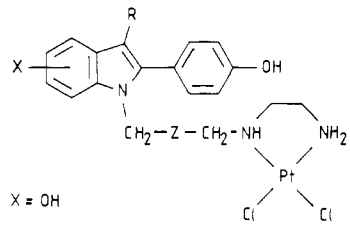
Table II. Dihydroxy-1-[[(2-aminoethyl)amino]alkyl]-2-phenylindoles and Their Estrogen Receptor Affinities



compd	R	position of X	Z	formula	mp,[a] °C	RBA[b]
10a	H	5	$(CH_2)_2$	$C_{20}H_{25}N_3O_2$	118–121	0.3
10b	H	6	$(CH_2)_2$	$C_{20}H_{25}N_3O_2$	99–101	0.04
10c	CH_3	5	$(CH_2)_2$	$C_{21}H_{27}N_3O_2$	145–147	1.0
11c	CH_3	5	$(CH_2)_3$	$C_{22}H_{29}N_3O_2$	126–128	1.9
12a	H	5	$(CH_2)_4$	$C_{22}H_{29}N_3O_2$	105	0.4
12b	H	6	$(CH_2)_4$	$C_{22}H_{29}N_3O_2$	149	0.3
12c	CH_3	5	$(CH_2)_4$	$C_{23}H_{31}N_3O_2$	112–114	10.0
13c	CH_3	5	C_6H_4	$C_{25}H_{27}N_3O_2$	165–167	11.6

[a] Crystallized from MeOH. [b] Relative binding affinities for the calf uterine estrogen receptor = ratio of molar concentrations of 17β-estradiol (E2) and inhibitor required to decrease the amount of bound [^{3}H]E2 by 50%, ×100.

Table III. [*N*-[ω-(Dihydroxy-2-phenylindol-1-yl)alkyl]-1,2-diaminoethane]dichloroplatinum(II) Complexes and Their Estrogen Receptor Affinities



compd	R	position of X	Z	formula[a]	dec,[b] °C	RBA[c]
14a	H	5	$(CH_2)_2$	$C_{20}H_{25}Cl_2N_3O_2Pt$[d]	>140	0.1
14b	H	6	$(CH_2)_2$	$C_{20}H_{25}Cl_2N_3O_2Pt$	>104	0.02
14c	CH_3	5	$(CH_2)_2$	$C_{21}H_{27}Cl_2N_3O_2Pt$[e]	>161	1.0
15c	CH_3	5	$(CH_2)_3$	$C_{22}H_{29}Cl_2N_3O_2Pt$	>127	1.3
16a	H	5	$(CH_2)_4$	$C_{22}H_{29}Cl_2N_3O_2Pt$	>155	0.5
16b	H	6	$(CH_2)_4$	$C_{22}H_{29}Cl_2N_3O_2Pt$	>125	0.5
16c	CH_3	5	$(CH_2)_4$	$C_{23}H_{31}Cl_2N_3O_2Pt$	>130	6.5
17c	CH_3	5	C_6H_4	$C_{25}H_{27}Cl_2N_3O_2Pt$[f]	>190	4.4

[a] Analyzed for C and H within ±0.5% of the calculated values, except where noted. [b] Crystallized from water. [c] Relative binding affinities for the calf uterine estrogen receptor = ratio of molar concentrations of 17β-estradiol (E2) and inhibitor, required to decrease the amount of bound [^{3}H]E2 by 50%, ×100. [d] C: calcd, 39.67; found, 38.09. [e] C: calcd, 40.71; found, 38.40. [f] C: calcd, 44.98; found, 43.99.

range from 20 to 50%. The melting points are reported in Table II.

General Procedure for the Preparation of the Dichloroplatinum(II) Complexes 14–17. A solution of K_2PtCl_4 (1.01 mmol) in 10 mL of a mixture of DMF and water (5:2, v/v) was added slowly to a warm (40 °C) solution of the (diaminoalkyl)-indole (1.01 mmol) in 10 mL of DMF. the mixture that exhibited a pH value of 9–10 was gently stirred in the dark at 30 °C for 3–5 days until the pH value reached 4–5. After addition of 1 drop of DMSO, stirring was continued for 2 h, followed by removal of the solvent in vacuo. The oily residue was suspended in saturated KCl solution, filtered, and washed with EtOH and water. For further purification, the product was dissolved in a small volume of DMF and precipitated with EtOH/water (1:1). After filtration the crystalline product was dried for several days. Sometimes the procedure for purification had to be repeated. All of the complexes do not melt but decompose at higher temperature (see Table III).

Estradiol Receptor Binding Assay. Reagents. [2,4,6,7-^{3}H]Estradiol (110 Ci/mmol) was obtained from New England Nuclear, Dreieich, FRG. Hormones and biochemicals were purchased from Sigma, München, FRG. TEA [Tris buffer (0.01 M, pH 7.5) supplemented with EDTA (0.01 M) and NaN_3 (0.003 M)] was used as buffer.

Fresh calf uteri, stored in ice-cold saline, were freed of adherent fat and connective tissue at 4 °C. After addition of TEA buffer (1 mL/g), the uteri were homogenzied by treatment with an ultraturrax mixer (IKA, FRG) and a glass-in-glass homogenizer (Potter S; Braun, FRG) at 4 °C. Lipids were separated by centrifugation at 700*g* and discarded. The homogenate was centrifuged at 105000*g* for 1 h (0 °C). The supernatant (cytosol) was then used for determining the affinity of compounds for the estrogen receptor. The protein concentration of the cytosol was ca. 15 mg/mL, leading to a final concentration of 3 mg/mL in the assay.

For the determination of the relative binding affinity (RBA), the previously described procedure was applied with modifications.[7] The 500-μL incubation mixture comprised 5 nM [^{3}H]-17β-estradiol (added in 100 μL of TEA), 10^{-9}–10^{-5} M competing ligand (in 100 μL of TEA), 100 μL of uterine cytosol, and TEA. The mixture was incubated for 18 h at 4 °C, and then 0.5 mL of dextran-coated charcoal (DCC) slurry (0.8% charcoal Norit A and 0.008% dextran in TEA) was added to the tubes, and the contents were mixed. The tubes were incubated for 90 min at 4 °C and then centrifuged at 700*g* for 10 min to pellet the charcoal. An aliquot (100 μL) of the supernatant was removed, and the radioactivity was determined by liquid scintillation spectrometry after addition of 2 mL of Quickszint 212 (Zinsser). Nonspecific binding was calculated with 5 μM 17β-estradiol as competing ligand. Six concentrations of competitor (1, 2, 5, and 10×10^{-9} to 10^{-6}) were chosen to provide values between 10 and 90% of specifically bound radioactivity. Radioactivity was plotted as a function of the log concentration of competing ligand in the assay. The RBA was calculated as the ratio of the molar concentrations of estradiol and test compound required to decrease the amount of bound radioactivity by 50%, multiplied by 100.

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Registry No. 1a, 5883-83-0; 1b, 62655-56-5; 1c, 91444-16-5; 2a, 115119-02-3; 2b, 115119-03-4; 2c, 115119-04-5; 3c, 115119-05-6; 4a, 115119-06-7; 4b, 115119-07-8; 4c, 115084-37-2; 5c, 115119-08-9; 6a, 115119-09-0; 6b, 115119-10-3; 6c, 115119-11-4; 7c, 115119-12-5; 8a, 115119-13-6; 8b, 115119-14-7; 8c, 115084-38-3; 9c, 115119-15-8; 10a, 115119-16-9; 10b, 115119-17-0; 10c, 115119-18-1; 11c, 115119-19-2; 12a, 115119-20-5; 12b, 115119-21-6; 12c, 115084-39-4; 13c, 115119-22-7; 14a, 115140-97-1; 14b, 115140-98-2; 14c, 115160-74-2; 15c, 115140-99-3; 16a, 115160-75-3; 16b, 115141-00-9; 16c, 115141-01-0; 17c, 115141-02-1; $Br(CH_2)_4Br$, 110-52-1; $Br(CH_2)_5Br$, 111-24-0; $Br(CH_2)_6Br$, 629-03-8; $BrCH_2C_6H_4$-p-CH_2Br, 623-24-5; K_2PtCl_4, 10025-99-7; $NH_2(CH_2)_2NH_2$, 107-15-3; 17β-estradiol, 50-28-2.

Supplementary Material Available: ^1H NMR data of 1-(bromoalkyl)-2-phenylindoles 2–5, 1-[[(2-aminoethyl)amino]alkyl]-2-phenylindoles 6–9, dihydroxy-1-[[(2-aminoethyl)amino]-alkyl]-2-phenylindoles 10–13, and N-[ω-(dihydroxy-2-phenylindol-1-yl)alkyl]-1,2-diaminoethane dichloroplatinum(II) complexes 14–17 (5 pages). Ordering information is given on any current masthead page.

Design of Rat Renin Inhibitory Peptides†

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Because several well-studied strains of rats manifest spontaneous hypertension, we set out to design a renin inhibitor suitable for use in this species. On the basis of the sequence of the renin substrate, a series of substrate analogue inhibitory peptides were synthesized by systematically modifying the P5, P3, P2, P1P1′, P2′, P3′, and P4′ positions. In assays against rat plasma renin, we found that modifications at the C-terminal segment have a marked influence on potency, and that a secondary butyl side chain at the P2′ position is important for obtaining optimal activity. The structure at the P3′ position, however, could vary considerably without significant effect. The steric effect of the P2 position was important; there an isopropyl side chain provided optimal binding between the inhibitor and the enzyme. At the P3 and P5 positions, potency appeared to depend on aromatic side chains. The effects at the P1P1′ position of the transition-state residue (3S,4S)-4-amino-3-hydroxy-6-methylheptanoic acid (statine) and its congeners (3S,4S)-4-amino-3-hydroxy-5-phenylpentanoic acid (AHPPA) and (3S,4S)-4-amino-3-hydroxy-5-cyclohexylpentanoic acid (ACHPA) were found to depend on the sequence of the C-terminal segment. For peptides with an unfavorable C-terminal segment (-Ile-Phe-NH_2), AHPPA and ACHPA resulted in a surprising retention of potency. For peptides with a favorable C-terminal segment (-Leu-Phe-NH_2), the effect of AHPPA was mild, even though ACHPA still significantly enhanced potency. The hypotensive and plasma renin inhibitory effects of three of the analogues were then studied in anesthetized sodium-depleted rats. One of the compounds, acetyl-His-Pro-Phe-Val-Statine-Leu-Phe-NH_2 (IC_{50} against rat plasma renin of 30 nM at pH 7.4), proved to be a potent hypotensive agent and a potentially useful probe for the study of the renin–angiotensin system in rats.

The renin–angiotensin system plays an important role in the maintenance of blood pressure and electrolyte balance.[1] Its precise contribution to the development of essential hypertension, however, is still not known. Because converting enzyme inhibitors (compounds in wide clinical use for the treatment of hypertension) also affect bradykinin[2] and prostaglandin,[3] they cannot be used as pharmacologic probes. For this reason potent and specific inhibitors of the renin–angiotensin system would help us understand a variety of clinical and experimental forms of the disease. In recent years transition-state substrate analogues have been shown to be effective inhibitors of primate renin, with IC_{50} values in the subnanomolar range.[4-9] Although some studies are being made of their hypotensive effect in vivo, little use has been made of these analogues in studies of the role of the renin–angiotensin system in the pathogenesis of hypertension.

Physiological studies in the primate are both difficult and expensive, and well-defined genetic models of hypertension have not been developed in this species. A potent inhibitor of rat renin, suitable for use in vivo, would be of great experimental value because of the considerable body of hypertensive studies in this species, the availability of a number of genetically determined models of spontaneous hypertension, and the relative abundance and low cost of these animals. However, because renins of various species differ considerably in both structure and substrate specificity, structural knowledge of primate renin inhibitors cannot be directly extrapolated to the design of rat renin inhibitors.

Recently a substrate analogue with a hydroxyethylene component at the scissile bond position was reported to have an IC_{50} value against rat plasma renin of 0.8 nM.[10]

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