Synthesis, Structure and Characterization of New Olivacine Derivatives and Their Platinum(II) Complexes

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Three unknown olivacine derivatives (L-OCH_3, L-OH, L-OAc) were synthesized according to a procedure that involves only a few steps, and used as ligands in the synthesis of neutral chelate platinum(II) complexes. The ligands, as well as their platinum complexes were spectroscopically characterized with IR, $^1H/^{13}C$ NMR and UV/Vis spectroscopic techniques. The solid state structures of two ligands (L-OCH_3, L-OAc) and one complex {[(L-OCH_3)$PtCl_2$]} were determined by X-ray diffraction. Molecular structure analysis of the ligands and the complex indicated that complexation resulted in a change in the conformation of the bipyridine fragment of the compounds studied and caused other interesting effects. Two crystallographically independent molecules of the complex were revealed in the measured crystal, with short intramolecular H···H distances. The structural and energetic aspects of the conformational behavior of the ligand L-OCH_3 and the complex [(L-OCH_3)$PtCl_2$] have been analyzed using semiempirical PM3 methods. In vitro antiproliferative activity was preliminarily estimated.

Introduction

The quest for new antitumor agents among the platinum complexes is still active and seems to be deeply grounded because of the fact that two compounds from this group, cisplatin and carboplatin, belong to the most effective and widely used platinum anticancer drugs. However, there are limitations to their applicability because of the high level of resistance observed in various tumors.[1] Moreover, these drugs are cross-resistant to each other which may be as a result of their similarity as they are both compounds of type $[Pt(NH_3)_2X_2]$ and have the same neutral ligands.

The in vivo cisplatin resistance may be caused by three major mechanisms. Firstly, the intracellular concentrations of the drug may be lower as a result of increased efflux or blocked uptake. Secondly, the drug can be inactivated through the quenching of the monofunctional Pt-DNA adducts before the formation of cross-links through reaction with metallothioneins, glutathione or other intracellular ligands. Finally, the removal of the drug-DNA adducts may occur through cell-based repair, which includes nucleotide excision repair.[2] The need to circumvent these processes has led to a search for analogs which avoid the cross-resistance but achieve a similar level of effectiveness. Considering that carboplatin does not fulfil such requirements, probably because of the formation of DNA adducts identical to those formed by cisplatin, one strategy to overcome cisplatin (and carboplatin) resistance would be to employ platinum complexes with bulky neutral ligands that form very different types of DNA lesions and inhibit nucleotide excision repair.[3]

In fact, in vitro studies of complexes that have at least one neutral ligand that is bulkier than NH_3, such as aminocyclohexane or pyridine derivatives, showed diminished cross-resistance to cisplatin.[4,5]

Moreover, it has also been suggested that the bulkier ligand may inhibit the reactivity of the platinum drug with cellular glutathione and other sulfur donors in vivo, and in this way diminish the concentration of GSH-platinum complexes. These complexes are actively transported out of cells and contribute to cisplatin efflux.[6,7]

If the mode of resistance to cisplatin is the efficient repair of specific adducts, then the other effective means of circumventing such resistance might be to prepare platinum complexes with ligands that have intercalative properties.[8,9] This led to the idea of using three new olivacine derivatives (L-OCH_3, L-OH, L-OAc, Figure 1) as ligands in the neutral platinum(II) complexes.

Olivacine is an isomeric form of ellipticine, which is a well-known cytostatic that acts through intercalation. Ellipticine is used as an anticancer drug in the form of elliptinium acetate (Figure 2, **NMHE**).[10,11] Recently, a series of new very promising antitumor olivacine derivatives, such as 9-hydroxy-5,6-dimethyl-6*H*-pyrido[4,3-*b*]carbazole-1-*N*-

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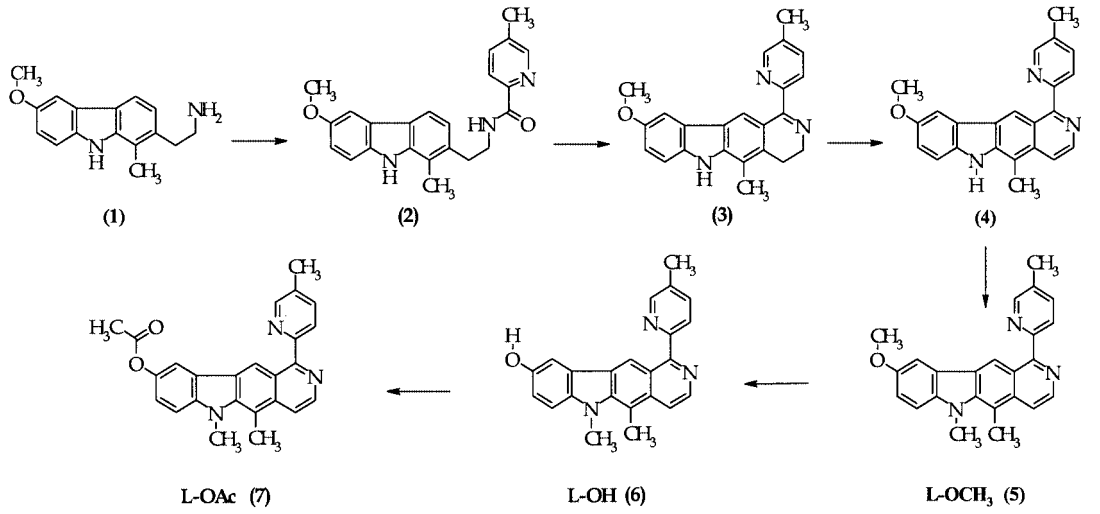

Figure 1. Synthetic pathway for the preparation of the ligands

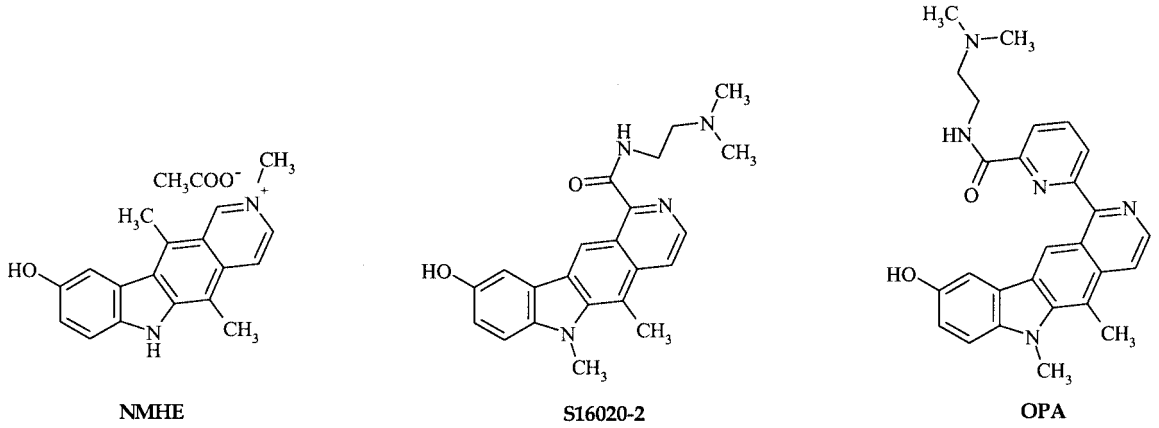

Figure 2. Ellipticine and olivacine derivatives: elliptinium acetate (**NMHE**); NSC-6596871 (**S16020−2**); olivacine picolinic amide (**OPA**)

(dimethylaminoethyl) carboxamide (Figure 2, **S16020−2**) and 6-(9-hydroxy-5,6-dimethyl)-6*H*-pyrido[4,3-*b*]carbazol-1-yl)picolinic (dimethylamino)ethylamide (Figure 2, **OPA**), has been synthesized and analyzed.[12−14]

The high antitumor activity of the above-mentioned olivacine derivatives encouraged us to search for new analogues which have already been preliminarily tested by us, and which have been shown to have significant cytotoxic activity in vitro.[15] In this paper, we present the synthesis of these analogues.

In Conclusion, we believe that the use of heterocyclic sterically hindered olivacine derivatives as ligands of metal complexes leads to a new group of effective platinum-containing cytostatics that would not be cross-resistant with the platinum-based drugs used so far.

Results and Discussion

Synthesis and Identification

Olivacine derivatives (L-OCH_3, L-OH, L-OAc) were synthesized as shown in Figure 1. The synthesis involved the use of 2-(6-methoxy-1-methylcarbazol-2-yl)ethylamine (**1**) as the starting compound, which was obtained according to an earlier procedure.[12] The reaction of **1** with 5-methyl-2-pyridinecarboxylic acid, and cyclization of the resulting new amide **2** using phosphorous oxychloride in boiling toluene, led to the formation of 9-methoxy-5-methyl-1-(5′-methylpyridin-2-yl)-3,4-dihydro-6*H*-pyrido[4,3-*b*]carbazole (**3**). This was aromatized to **4** by dehydrogenation over 10% palladium on charcoal in boiling diphenyl ether. *N*-6-methylation of **4** to (L-OCH_3) was then performed using an excess of dimethyl carbonate in dimethyl formamide, in the presence of potassium carbonate and 18-crown-6. Compound (L-OCH_3) was 9-*O*-demethylated to give 9-hydroxy-5,6-dimethyl-1-(5′-methylpyridin-2-yl)-6*H*-pyrido[4,3-*b*]carbazole (L-OH), by heating with a 48% aqueous solution of hydrobromic acid. The 9-hydroxy derivative, (L-OH), was allowed to react with a mixture of acetic anhydride/pyridine (1:1) at room temperature. This led to the formation of 9-acetoxy-5,6-dimethyl-1-(5′-methylpyridin-2-yl)-6*H*-pyrido[4,3-*b*]carbazole (L-OAc).

The new platinum(II) complexes, [(L-OCH_3)$PtCl_2$], [(L-OH)$PtCl_2$], [(L-OAc)$PtCl_2$], were prepared from K_2PtCl_4

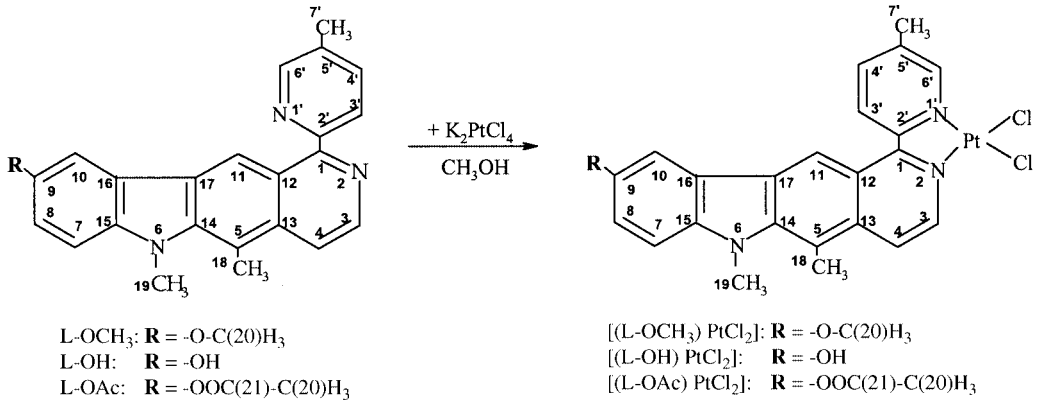

Figure 3. Scheme showing the formation of the complexes and the numbered atoms

and the olivacine derivatives, L-OCH_3, L-OH and L-OAc (Figure 3), respectively, according to a modified version of previously published methods.[16,17]

Chemical analyses, TLC, and spectroscopic techniques [MS (*m*/*z*), ^{13}C and/or ^{1}H NMR, UV/Vis, IR] were used for the characterization of the olivacine derivatives, as well as their platinum complexes. The platinum complexes were characterized additionally using electric conductivity measurements, which showed their neutral character. On the basis of the above physico-chemical studies, the molecular formulae of all the investigated compounds were postulated. The structures of the two ligands (L-OAc, L-OCH_3) and one platinum complex {[(L-OCH_3)$PtCl_2$]} were established by X-ray analysis. The structural and energetic aspects of the conformational behavior of the ligand/complex system have been analyzed.

FT-IR Characterization

The bands were assigned using literature data for the vibrational spectra of pyridine and its derivatives,[18−21] bipyridine,[22] carbazole derivatives [23] and platinum complexes.[24−35] Despite the general similarity of the spectra of the ligands, a few significant differences were noticed. Namely, only the IR spectrum of L-OH contains peaks that are characteristic for ν(C-OH) (at 1235 cm^{-1}) and for δ(O−H) (doublet at 852 and 843 cm^{-1} and band at 665 cm^{-1}). The IR spectrum of L-OAc shows a characteristic strong band at 1753 cm^{-1}, which is assigned to ν(C=O) of the 9-($OOCCH_3$) substituent, whereas the in the IR spectrum of L-OCH_3, a band at 2824 cm^{-1} is assigned to ν_s(C−H) of the OCH_3 group.

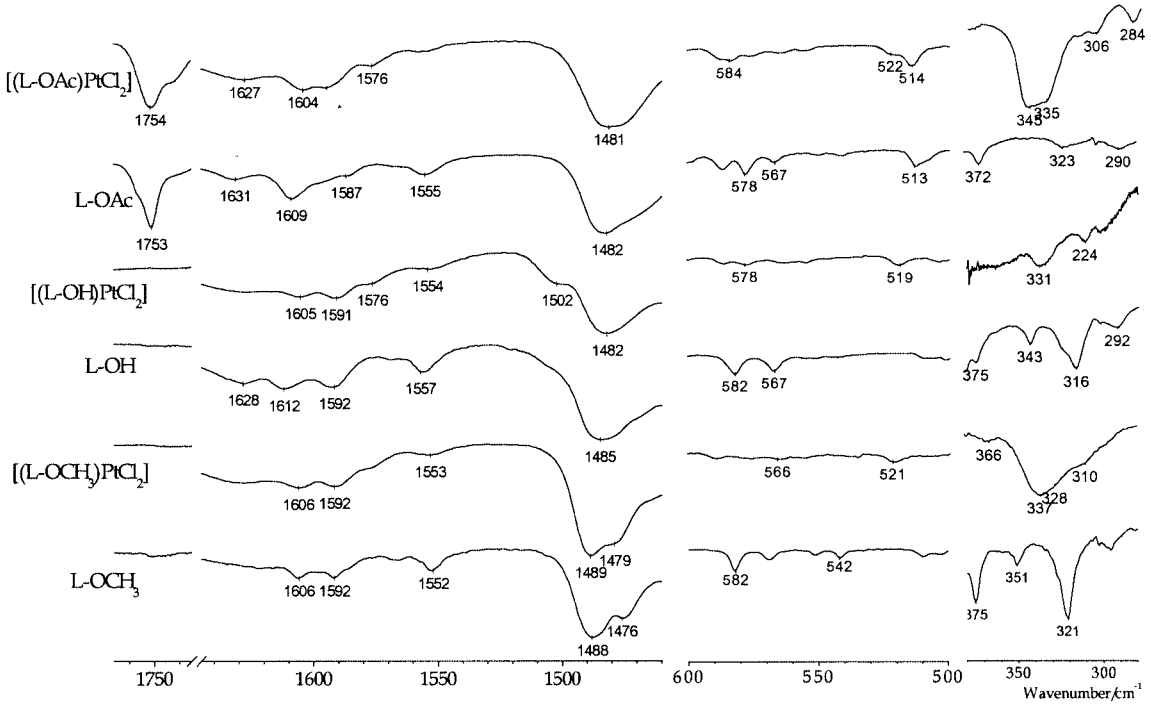

Figure 4. IR spectra of the ligands and Pt complexes

After complexation, changes in the position and intensity of some of the above bands and the appearance of new bands are observed (Figure 4). The new bands in the complexes that appear in the range 514−521 cm^{-1} and 330−350 cm^{-1} are assigned to ν(py) + ν(Pt−N) and ν(Pt−Cl), respectively. We can also identify the new bands at around 300 cm^{-1} and below; they seem to occur as a result of the bending vibrations of the N−Pt−N and/or N−Pt−Cl bonds. In general, the IR results show that the N(1′) and N(2) atoms donate electron density to the Pt^{2+} ion that is simultaneously bonded to the Cl anions.

NMR Spectroscopic Characterization

The ^{1}H NMR spectroscopic data for the 9-olivacine derivatives (ligands) and their platinum complexes are listed in Table 1. The ^{13}C NMR spectroscopic data, which were gathered only for the ligands, are presented in Table 2. The atom (H and C) numbering is according to Figure 3. The ^{1}H NMR spectra of the 9-olivacine derivatives were recorded using CD_2Cl_2 and [D_7]DMF as solvents. CD_2Cl_2 was the more convenient solvent since its signals does not overlap those of the olivacine derivatives. However, the Pt

Table 1. Chemical shift values (δ [ppm]), integrals, and coupling constants (*J* [Hz]) in the ^{1}H NMR spectra of the ligands and their platinum complexes

	−**CH**$_3$				−**CH**								
Proton's number	7′	18	19	20	3	4	7	8	10	11	3′	4′	6′
L-OCH_3 (CD_2Cl_2)	2.49 s, 3H	3.12 s, 3H	4.13 s, 3H	3.89 s, 3H	8.52 d, 1H $J_{3,4}$ = 6.4	7.97 d, 1H $J_{4,3}$ = 6.2	7.31 d, 1H $J_{7,8}$ = 8.7	7.17 dd, 1H $J_{8,7}$ = 8.7 $J_{8,10}$ = 2.5	7.60 d, 1H $J_{10,8}$ = 2.5	9.15 s, 1H	7.90 d, 1H $J_{3',4'}$ = 8.2	7.75 dd, 1H $J_{4',3'}$ = 8.2 $J_{4',6'}$ = 1.9	8.69 s, 1H
L-OH (CD_2Cl_2)	2.43 s, 3H	3.08 s, 3H	4.05 s, 3H	−	8.51 d, 1H $J_{3,4}$ = 6.2	7.97 d, 1H $J_{4,3}$ = 6.2	7.12 d, 1H $J_{7,8}$ = 8.6	6.89 dd, 1H $J_{8,7}$ = 8.5 $J_{8,10}$ = 2.3	7.49 d, 1H $J_{10,8}$ = 2.2	9.00 s, 1H	7.91 d, 1H $J_{3',4'}$ = 7.8	7.76 dd, 1H $J_{4',3'}$ = 7.8 $J_{4',6'}$ = 1.5	8.61 s, 1H
L-OAc (CD_2Cl_2)	2.49 s, 3H	3.13 s, 3H	4.16 s, 3H	2.32 s, 3H	8.54 d, 1H $J_{3,4}$ = 6.3	7.98 d, 1H $J_{4,3}$ = 6.3	7.37 d, 1H $J_{7,8}$ = 8.6	7.25 dd, 1H $J_{8,7}$ = 8.7 $J_{8,10}$ = 2.4	7.80 d, 1H $J_{10,8}$ = 2.2	9.17 s, 1H	7.90 d, 1H $J_{3',4'}$ = 7.8	7.74 dd, 1H $J_{4',3'}$ = 7.8 $J_{4',6'}$ = 2.2	8.67 s, 1H
L-OCH_3 ([D_7]DMF)	2.52 s, 3H	3.21 s, 3H	4.27 s, 3H	3.94 s, 3H	8.58 d, 1H $J_{3,4}$ = 6.2	8.17 d, 1H $J_{4,3}$ = 6.2	7.61 d, 1H $J_{7,8}$ = 8.9	7.27 dd, 1H $J_{8,7}$ = 8.6 $J_{8,10}$ = 2.8	7.89 d, 1H $J_{10,8}$ = 2.6	9.43 s, 1H	8.03 d, 1H partly covered	7.94 dd, 1H $J_{4',3'}$ = 7.9 $J_{4',6'}$ = 1.4	8.72 s, 1H
[(L-OCH_3)$PtCl_2$] ([D_7]DMF)	2.55 s, 3H	3.29 s, 3H	4.39 s, 3H	3.99 s, 3H	8.30 d, 1H $J_{3,4}$ = 8.5	8.24 d, 1H $J_{4,3}$ = 8.4	7.71 d, 1H $J_{7,8}$ = 8.9	7.38 dd, 1H $J_{8,7}$ = 8.8 $J_{8,10}$ = 2.6	8.16 d, 1H $J_{10,8}$ = 2.5	9.51 s, 1H	9.35 d, 1H $J_{3',4'}$ = 7.0	8.36 d, 1H $J_{4',3'}$ = 7.1	9.46 s, 1H
L-OH ([D_7]DMF)	2.53 s, 3H	3.18 s, 3H	4.22 s, 3H	−	8.57 d, 1H $J_{3,4}$ = 4.7	8.13 d, 1H $J_{4,3}$ = 5.9	7.50 d, 1H $J_{7,8}$ = 8.7	7.19 dd, 1H $J_{8,7}$ = 8.7 $J_{8,10}$ = 2.4	7.65 d, 1H $J_{10,8}$ = 2.4	9.33 s, 1H	d, 1H partly covered	7.94 dd, 1H $J_{4',3'}$ = 7.9 $J_{4',6'}$ = 1.2	8.74 s, 1H
[(L-OH)$PtCl_2$] ([D_7]DMF	2.57 s, 3H	3.25 s, 3H	4.33 s, 3H	−	8.81 d, 1H $J_{3,4}$ = 8.5	8.26 d, 1H $J_{4,3}$ = 7.5	7.59 d, 1H $J_{7,8}$ = 8.9	7.31 dd, 1H $J_{8,10}$ = 2.3	7,92 s, 1H	9.40 s, 1H	9.32 d, 1H $J_{3',4'}$ = 6.9	8.31 d, 1H $J_{4',3'}$ = 6.9	9.35 s, 1H
L-OAc ([D_7]DMF)	2.50 s, 3H	3.15 s, 3H	4.22 s, 3H	2.35 s, 3H	8.60 s, 1H	8.19 d, 1H $J_{4,3}$ = 5.1	7.66 d, 1H $J_{7,8}$ = 8.8	7.37 dd, 1H $J_{8,7}$ = 8.7 $J_{8,10}$ = 2.3	8.01 d, 1H $J_{10,8}$ = 2.3	9.42 s, 1H	covered	7.91 d, 1H $J_{4',3'}$ = 7.9	8.73 s, 1H
[(L-OAc)$PtCl_2$] ([D_7]DMF)	2.46 s, 3H	3.23 s, 3H	4.35 s, 3H	2.45 s, 3H	8.90 d, 1H $J_{3,4}$ = 5.2	8.08 d, 1H $J_{4,3}$ = 5.6	7.74 d, 1H $J_{7,8}$ = 9.0	7.49 d, 1H $J_{8,7}$ = 8.5	8.45 s, 1H	9.57 s, 1H	9.35 s, 1H	8.24 s, 1H	9.46 s, 1H

Table 2. Chemical shift values (δ [ppm]) in the ^{13}C NMR spectra of the ligands

Carbon's number	1	3	4	5	7	8	9	10	11	12	13	16	18	19	20	21	2′	3′	4′	5′	6′	7′
L-OCH_3 (CD_2Cl_2)	158.82	140.49	116.36	111.55	109.69	116.40	154.41	104.49	117.73	135.68	121.64	126.67	14.15	34.12	56.33	−	157.10	125.16	137.49	132.97	149.22	18.46
L-OH (CD_2Cl_2)	158.08	141.24	116.60	111.80	109.63	116.57	150.58	106.88	117.75	135.63	121.12	126.56	14.06	33.85	−	−	156.59	125.44	137.77	133.33	148.63	18.41
L-OAc (CD_2Cl_2)	158.83	140.68	116.45	111.89	109.23	121.50	142.96	113.86	118.23	135.93	121.83	126.15	14.10	34.12	21.21	170.42	156.91	125.15	137.50	133.06	149.17	18.44

complexes are sparingly soluble in CD_2Cl_2, hence their ^1H NMR spectra were recorded in [D_7]DMF. The use of two solvents was intentional since it allowed us to assign all the proton chemical shift values of the ligands, and in this way help in the analysis of the ^1H NMR spectra of the Pt complexes. The ^{13}C NMR spectra could be recorded only for the ligands since the Pt complexes were relatively insoluble in all the solvents used.

The assignments of the ^1H and ^{13}C NMR chemical shifts for the ligands were completed using two-dimensional HMQC, HMBC and COSY techniques; however, for the Pt complexes, only the ^1H NMR peaks could be assigned by comparing the spectra of the complexes with that of the corresponding ligands, and using data from the literature.[12,14] The proton chemical shift values for compounds **2**, **3** and **4** were measured and the data is presented in the Exp. Sect.

There is an appreciable downfield shift of almost all the signals in the ^1H NMR spectra of the Pt complexes relative to those in the spectra of the free ligands. However, there is a greater downfield shift of the signals for the protons at position 3′, 4′ and 6′. This is in agreement with IR results and leads to the conclusion that ligands exhibit a neutral bidentate function through the N2 and N1′ atoms. The downfield shift of the signals for the protons in neighboring region of the nitrogen atoms that are coordinated to the platinum ion has been reported for other chelate platinum complexes with nitrogen ligands.[36−38]

UV/Vis Characterization

Data obtained from the electronic spectra of the ligands (L-OCH_3, L-OH, L-OAc) and their Pt complexes ([(L-OCH_3)$PtCl_2$], [(L-OH)$PtCl_2$], [(L-OAc)$PtCl_2$]) in methanol and chlorobenzene solutions are presented in Table 3.

The UV/Vis bands of the ligands and Pt complexes were assigned using data from the literature for free bipyridine derivatives and their Pt complexes.[39−47] The authors in the above-mentioned references usually observed two bands, which were assigned as charge transfer transitions from the platinum d-orbital to π-anti-bonding orbitals of the bipyridyl group (MLCT); the energies were strongly dependent on the identity of the other ligands coordinated to the platinum ion and the solvent (e.g. in water, the first MLCT band appeared at 345 nm, whereas in chlorobenzene the band was at 413 nm).[39] In our studies on platinum complexes, in methanol, the band appears between 420−428 nm, and in chlorobenzene, the band appears at 440 nm. These were assigned to the first MLCT. This absorption band, which is correlated to the polarity of solvents, must be due to the platinum d → bipyridyl π* transition.

The second MLCT band in the spectra of our complexes also show a solvent dependency; in methanol, the band appears between 272−278 nm, whereas in chlorobenzene, the band is shifted to 288−292 nm.

According to the data published for other bipyridine platinum complexes, we assigned the intensive band in the

Table 3. Data from the UV/Vis measurements of the ligands and their platinum complexes in two solvents

Wavelength [nm] (ε [$mol^{-1}\cdot dm^3\cdot cm^{-1}$])						Assignments
L-OCH_3	[(L-OCH_3)$PtCl_2$]	L-OH	[(L-OH)$PtCl_2$]	L-OAc	[(L-OAc)$PtCl_2$]	
212[a]	210[a]	212[a]	208[a]	210[a]	210[a]	
($1.1\cdot10^5$)	($1.7\cdot10^4$)	($8.4\cdot10^4$)	($1.7\cdot10^4$)	($8.6\cdot10^4$)	($3.7\cdot10^4$)	
solvent abs.[b]	solvent abs.[b]	solvent abs.[b]	solvent abs.[b]	solvent abs.[b]	solvent abs.[b]	π → π*
274[a]		274[a]		272[a]		(intra ligand)
($1.4\cdot10^5$)		($8.7\cdot10^4$)		($1.2\cdot10^5$)		
solvent abs.[b]		solvent abs.[b]				
	278[a]		272[a]		278[a]	MLCT
	($1.3\cdot10^4$)		($1.1\cdot10^4$)		($3.2\cdot10^4$)	(spin
	292[b]		290[b]		288[b]	forbidden)
	($2.0\cdot10^4$)		($1.1\cdot10^4$)		($1.8\cdot10^4$)	
306[a]		308[a]		302[a]		
($2.0\cdot10^5$)		($1.2\cdot10^5$)		($2.1\cdot10^5$)		
312		312[b]		306[b]		
($1.5\cdot10^5$)		($3.9\cdot10^4$)		($9.83\cdot10^4$)		π → π*
	350[a]		348[a]		342[a]	(intra ligand)
	($1.3\cdot10^4$)		($8.7\cdot10^3$)		($3.3\cdot10^4$)	
	356[b]		354[b]		348[b]	
	($2.6\cdot10^4$)		($6.7\cdot10^3$)		($2.8\cdot10^4$)	
	420[a]		428[a]		420[a]	MLCT
	($6.7\cdot10^3$)		($3.8\cdot10^3$)		($1.4\cdot10^4$)	(spin
	440[b]		440[b]		440[b]	allowed)
	($7.7\cdot10^3$)		($2.0\cdot10^3$)		($6.7\cdot10^3$)	

[a] Methanol solution. [b] Chlorobenzene solution.

Table 4. Crystal data and structure refinement for **5**, **7** and **8**

Compound	L-OCH_3 (**5**)	L-OAc (**7**)	[(L-OCH_3)$PtCl_2$] (**8**)
CCDC no.	203648	203649	203650
Empirical formula	$C_{24}H_{21}N_3O$	$C_{25}H_{21}N_3O_2$	$C_{24}H_{21}Cl_2N_3OPt \times C_3H_7NO \times 0.25H_2O$
Formula mass	367.44	395.45	711.03
Crystal system	monoclinic	monoclinic	monoclinic
Space group	$P2_1/c$	$P2_1/n$	$P2_1/n$
a (Å)	7.175(3)	14.369(3)	14.233(3)
b (Å)	20.072(7)	8.050(2)	25.569(4)
c (Å)	13.105(5)	17.074(3)	14.700(3)
β [°]	101.51(4)	96.18(2)	107.83(3)
V (Å^3)	1849.4(12)	1963.5(7)	5092.7(17)
Z	4	4	8
$d_{calcd.}$ [g·cm^{-3}]	1.320	1.338	1.855
T [K]	100	100	100
μ [mm^{-1}]	0.082	0.086	5.76
F(000)	776	832	2788
Crystal size [mm]	0.8 × 0.1 × 0.06	0.6 × 0.2 × 0.1	0.06 × 0.05 × 0.02
θ range [°]	3.5 to 28.5	3.5 to 28.7	3.3 to 28.5
Reflections measured	11673	12818	29166
Unique reflections ($R_{int.}$)	4318 (0.0721)	4643 (0.0265)	11396 (0.1085)
Absorptio correction	none	none	analytical
Transmission, max./min.			0.904/0.695
Extinction	0.0121(15)		
Parameters/constraints	317/0	355/0	349/27
R_1 [$I > 2\sigma(I)$]	0.0590	0.0396	0.0699
Data with $I > 2\sigma(I)$	2897	3615	6015
R_1(all data)	0.1086	0.0570	0.1769
wR_2(all data, on F^2)	0.1208	0.1044	0.1048
S	1.039	1.095	0.874
Residual density [e·Å^{-3}]	−0.26/0.22	−0.22/0.20	−1.63/1.18

Table 5. Shortest intra- and intermolecular hydrogen bonds for **5**, **7**, and **8**, with standard deviations in parentheses; square brackets denote short contacts for disordered atoms; symmetry codes: i ($x-1, y, z$); ii ($2-x, 1-y, 1-z$); iii ($1+x, 0.5-y, z-0.5$); iv ($0.5+x, 0.5-y, z-0.5$); v ($x-0.5, 0.5-y, 0.5+z$); vi ($1.5-x, 0.5+y, 1.5-z$); vii ($x+0.5, -y+0.5, z+0.5$); viii ($x-0.5, 0.5-y, z-0.5$)

Compound	D−H···A	D···A [Å]	<(DHA) [°]
L-OCH_3 (**5**)	C(11)−H(11)···N(18)	2.976(3)	115.6(13)
	C(20)−H(20)···N(2)	2.820(3)	94.5(13)
	C(23)−H(23)···N(2)^{i}	3.445(3)	150.8(14)
	C(20)−H(20)···C(9)^{ii}	3.412(3)	115.3(14)
	C(3)−H(3)···C(01)^{iii}	3.582(3)	138.9(15)
	C(92)−H(922)···C(12)^{i}	3.801(3)	162.8(15)
	C(92)−H(922)···C(13)^{i}	3.814(3)	149.3(15)
L-OAc (**7**)	C(11)−H(11)···N(18)	2.917(2)	120.7(10)
	C(20)−H(20)···N(2)	2.810(2)	96.5(9)
	C(21)−H(21)···O(94)^{iv}	3.330(2)	124.7(11)
	C(8)−H(8)···N(2)^{v}	3.544(2)	161.6(11)
	C(93)−H(933)···N(18)^{vi}	3.409(2)	134.0(13)
[(L-OCH_3)$PtCl_2$] (**8**)	C(3a)−H(3a)···Cl(2a)	3.193(12)	123
	C(23a)−H(23a)···Cl(1a)	3.253(12)	122
	[C(21a)−H(21a)···O(1e)]	3.14(2)	154
	[C(21a)−H(21a)···O(1d)]	3.24(2)	156
	C(3b)−H(3b)···Cl(2b)	3.207(13)	120
	C(23b)−H(23b)···Cl(1b)	3.299(11)	121
	[C(10b)−H(10b)···O(1d)]^{vii}	3.40(2)	130
	[C(20b)−H(20b)···O(1d)]^{vii}	3.44(2)	132
	[C(20b)−H(20b)···O(1e)]^{vii}	3.15(2)	128
	[C(21b)−H(21b)···O(1c)]^{vii}	3.38(2)	136
	[O(1f)−H(1f1)···O(1c)]	2.70(2)	116
	O(1f)−H(1f2)···Cl(2a)^{viii}	3.78(2)	139

spectra of the ligands at 306 nm to an internal bipyridine ring transition.[38−40] In complexes [(L-OCH_3)$PtCl_2$], [(L-OH)$PtCl_2$] and [(L-OAc)$PtCl_2$], this band is bathochromic shifted by about 40 nm. Such a shift is consistent with reports in the literature on complexation or reduction of bipyridines.[39,46−50]

X-ray Structure

Details of the crystal parameters, data collection and structure refinement for L-OCH_3 (**5**), L-OAc (**7**) and [(L-OCH_3)$PtCl_2$] × DMF × 0.25H_2O (**8**), are given in Table 4. Selected hydrogen interactions are listed in Table 5.

The asymmetric unit of **8** contains two crystallographically independent complex molecules [(L-OCH_3)$PtCl_2$] (hereafter referred to as **a** and **b**), two molecules of DMF (one is disordered) and half a molecule of water.

Both molecular structures of ligands **5** and **7** (presented in Figure 5 and 6) reveal the intramolecular hydrogen bond between C(11)−H(11) and N(18), and a fairly short contact between C(20)−H(20) and N(2). The picoline ring in both ligands is twisted from the core formed by four other rings, with dihedral angles of 38.0(1)° and 36.3(1)° in **5** and **7**, respectively.

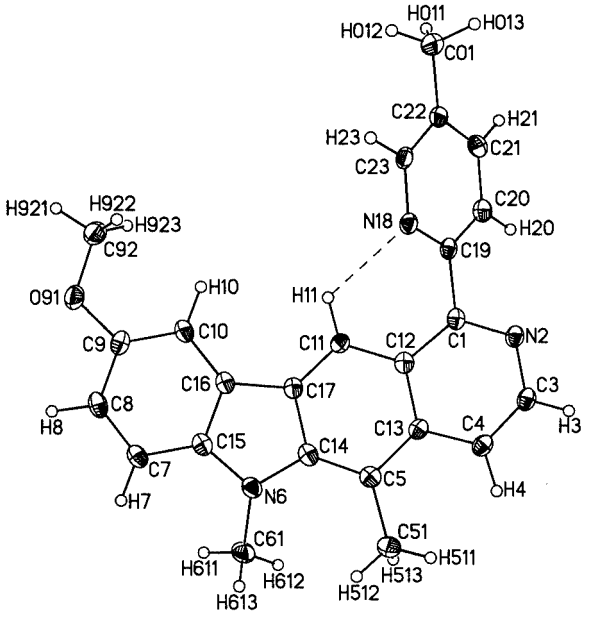

Figure 5. Molecular structure of **5**, showing the atom numbering scheme and displacement ellipsoid plotted at the 50% probability; four fused rings of the molecular core are in the plane of the sheet; the shortest intramolecular C−H···N hydrogen bond is shown as a dashed line

Analysis of the crystal packing reveals stacking interactions between consecutive parallel molecules; these are often observed for compounds with extended π-systems.[51] In the crystal cells of both ligands, columnar structures of the molecules are formed with the nearest neighbors, with a head-to-head orientation, and the distances between them allow for the C···C and C···H and N···H stacking interactions. In both ligands, mutual arene−arene interactions are

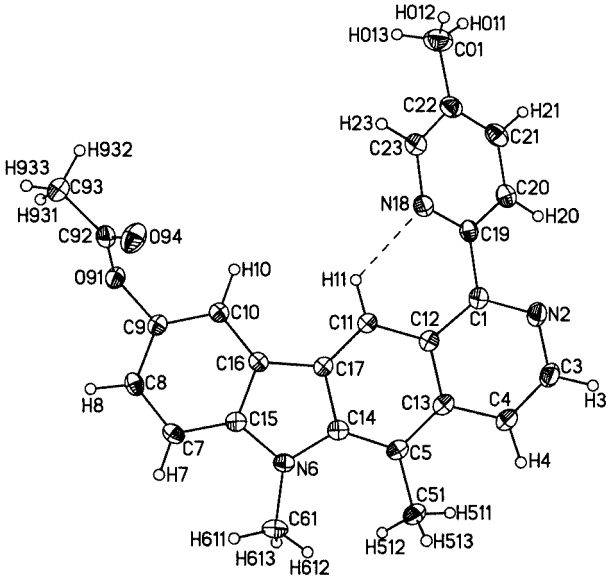

Figure 6. Molecular structure of **7**, showing the atom numbering scheme and displacement ellipsoid plotted at the 50% probability; four fused rings of the molecular core are in the plane of the sheet; the shortest intramolecular C−H···N hydrogen bond is shown as a dashed line

also observed through weak C−H···C and C−H···N intermolecular hydrogen bonds. However, as shown in Figure 7 and 8, some differences are seen in the crystal packing. The adjacent columns of bow-like shaped molecules of **5** are in a head-to-tail nonparallel mutual arrangement, and intermolecular hydrogen bonds are observed between them. In the crystal cell of **7**, the adjacent columns are oriented in an almost perpendicular manner.

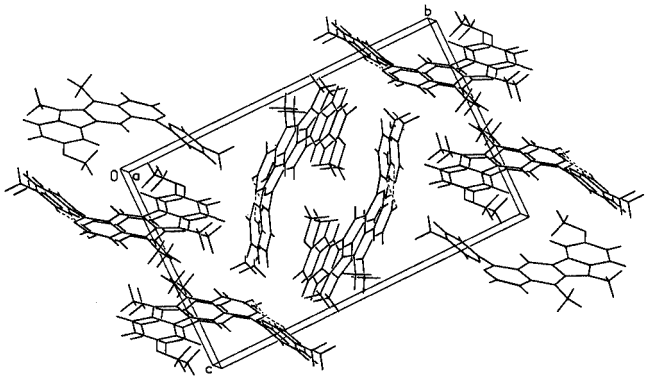

Figure 7. Crystal packing diagram for **5** along axis *a*

The relatively planar molecular structure and stacking interactions seen in **5** and **7** meet the structural requirements for the intercalative type of biological activity mechanism predicted for this type of compounds.[52]

The molecular structure of complex **8** obtained by X-ray diffraction unequivocally confirms that the bipyridine fragment is chelated to the platinum center; the remaining coordination sites are occupied by two *cis* chloride ligands. In molecules **a** and **b**, the coordination geometry around the platinum center is square planar. Although molecules **a** and

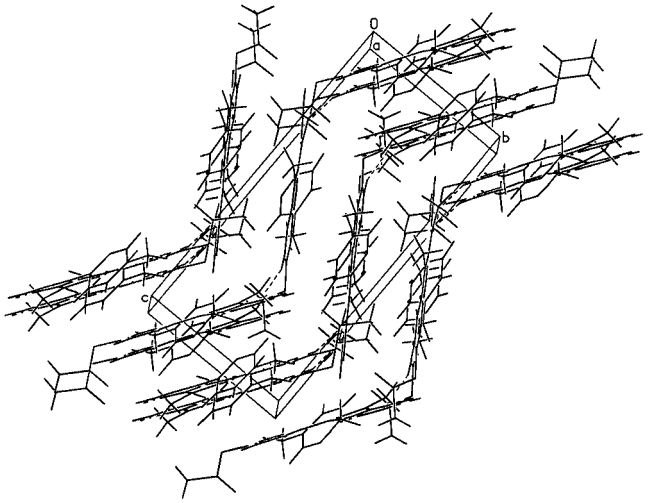

Figure 8. Crystal packing diagram for **7** along axis *a*

b are very similar, as seen in Figure 9, some differences can be observed.

Firstly, the methoxy substituent in molecule **b**, as in **5**, is in a *cis* configuration relative to C(10)−H(10), whereas in **a**, it is in a *trans* configuration.

Another difference is related to the value of dihedral angle between the picoline ring and the skeleton of the molecule (formed by four other rings); 13.9(3)° in **a**, and 16.7(3)° in **b**. This is further related to the proximity of the H(11) and H(20) atoms. Complexation resulted in picoline ring rotation, thus this distance is 1.67 in **a** and 1.88 Å in **b** (with the C−H distance assumed as 1.08 Å). The geometric effect forced by coordination and/or the crystal packing effect cannot be excluded as a reason for achieving such a short contact between H(11) and H(20) and for the appearance of two independent molecules observed in the crystalline state of **8**.

A comparison of the bond lengths reveals the next difference between **a** and **b** (and **5**). The differences in the bond lengths may be due to the change in the conjugation of the π-bonds, and might be related to the above-mentioned H(11)···H(20) interaction.

The Pt−N bond lengths in molecules **a** and **b** are similar to those in other chloro-platinum complexes with bisisoquinoline,[53] as well as with bipyridines.[35,54,55] In molecule **a**, both Pt−Cl bond lengths are within the expected range, whereas in molecule **b**, the Pt−Cl(1) bond is slightly longer [2.315(3) Å]. The chloride ligands are also engaged in weak intramolecular hydrogen bonds with C(23)−H(23) and C(3)−H(3).

The mode of π-π stacking interactions in the unit cell of **8** is more complicated than in **5**. Molecules **a** and **b** show alternative coordination modes of *cis*-[PtN_2Cl_2]. Consecutive molecules of **a** and **b** are not parallel and overlap each other, either with through the bipyridine fragments or through the "tails" of the molecules.

C···C and H···C stacking interactions are observed. Furthermore, N(6)···Pt and Pt···C(14) interactions can also be seen . In contrast to some other Pt complexes with similarly coordinated bipyridine ligands, there is no linear stacking

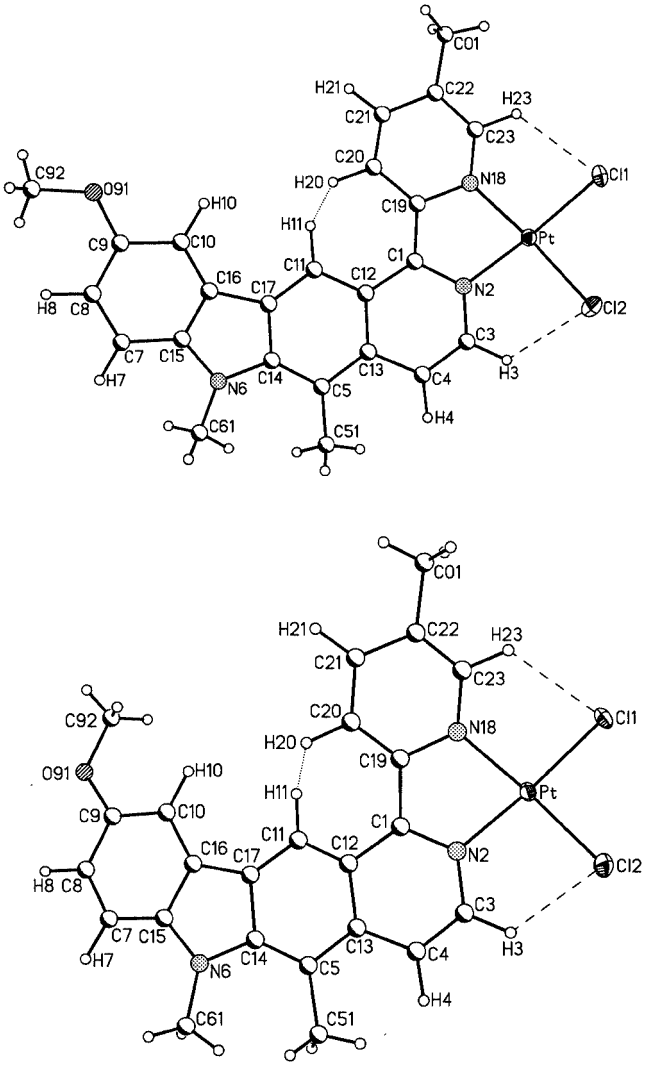

Figure 9. Molecular structure of **8 a** and **8 b**, showing the atom numbering scheme and displacement ellipsoid plotted at the 50% probability for the platinum center and the chloride ligands; the coordination plane defined by [PtN_2Cl_2] is in the plane of the sheet; the intramolecular C−H···Cl hydrogen bonds are shown as dashed lines; the shortest intramolecular distances between the hydrogen atoms are shown as a dotted line

arrangement of the platinum centers.[54] The shortest distance between the platinum centers in **a** and **b** is 6.2 Å, and suggests no significant Pt−Pt interaction (relative to the reported value 3.45 Å in ref.[54]).

Conformational Analysis of L-OCH_3

Due to symmetry, two conformers of the ligand L-OCH_3 can exist in a global energy minimum in the one molecule model gas state, with calculated torsion angles N(2)−C(1)−C(19)−N(18) of approximately −91° or 93,° respectively, for conformers in an energy minimum. The calculated binding energy of these conformers is −5537.71 kcal/mol (Table 6). The calculated standard enthalpy is 56.35 kcal/mol. In the crystalline state, the measured torsion angle N(2)−C(1)−C(19)−N(18) is −141.5(2)°, so the difference between the calculated torsion angle

Table 6. Calculated binding energies, heats of formation and energy barriers for rotation for the conformers of L-OCH_3

Conformers of L-OCH_3	Respected torsion angle N(2)−C(1)−C(19)−N(18)[a] [°]	Binding energy [kcal/mol]	Standard heat (enthalpy) of formation ΔH^{298} [kcal/mol]	Rotational barrier E_{rot} [kcal/mol]
In energy minimum	−91.16 or 92.64[b]	−5537.71	56.35	0.00
Antiplanar	180.0 (−180.0)	−5535.60	58.46	2.11
Synplanar	0.00	−5511.37	82.69	26.34
in [(L-OCH_3)$PtCl_2$] molecule **a**	−11.7(14)[c]	−5531.32	62.74	6.39

[a] Atom numbering according to that used for the X-ray structure. [b] Calculated. [c] From X-ray diffraction measurements.

N(2)−C(1)−C(19)−N(18) in the model gas state and that measured in the crystals is −50.49°. The observed difference might be caused by the fact that intermolecular interactions (e.g. electrostatic, Van der Waals, dispersion, hydrogen bonds) that exist in the real crystalline state, are not taken into consideration in the calculated method. The estimated binding energy for one of the planar conformers, for which N(2)−C(1)−C(19)−N(18) is 180° (or −180°), is −5535.60 kcal/mol, and the standard enthalpy is 58.46 kcal/mol. Moreover, the energy barrier for rotation around the C(1)−C(19) bond (when the torsion angle N(2)−C(1)−C(19)−N(18) changes from −91° to 180°), calculated from the difference in the enthalpies of formation of the proper conformers, is : E_{rot} = 2.11 kcal/mol (E_{rot} = ΔH_{rot} rotational enthalpy). This low energy barrier indicates that the antiplanar conformation of L-OCH_3 is easily achieved, and this may also explain the significant difference between the measured and calculated torsion angles because this fragment of the molecule is flexible enough [in this range for the torsion angle N(2)−C(1)−C(19)−N(18)], although rotation around the C(1)−C(19) bond is not completely free since the energy required is higher than the thermal energy (translation) at 298 K ($E_{rot} > RT$ = 0.592 kcal/mol). The energy barrier for rotation around the C(1)−C(19) bond (torsion angle N(2)−C(1)−C(19)−N(18) changes from −91° or 92° to 0°) in order to adopt a synplanar conformation is very high, E_{rot} = 26.34 kcal/mol (= rotational enthalpy), so this conformation is almost impossible to achieve at standard temperature, hence there is absolutely no rotation around this bond. If the Pt^{2+} ion required a synplanar [torsion angle N(2)−C(1)−C(19)−N(18) = 0°] conformation of the ligand L-OCH_3 for complexation, complex formation would probably not occur. However, according to the literature, the known complexes of Pt^{2+} with 1,1′-biisoquinoline[53] and 2,2′-bipyridines[56] have analogous torsion angles that are not planar. X-ray diffraction measurements show that in the case of [(1,1′-biisoquinoline)$PtCl_2$], these angles are 35.18° and 41.12°, and thus the molecule is very twisted.[53] In the case of unsubstituted [(2,2′-bipyridine)$PtCl_2$], the torsion angle indicated an almost planar molecule and the torsion angle was estimated to be 4.8(6)°, but 3,3′-bisubstituted [(2,2′-bipyridine)$PtCl_2$] is markedly non-planar [25.5(3)°].[56] The measured torsion angle N(2)−C(1)−C(19)−N(18) in the crystal of [(L-OCH_3)$PtCl_2$] for molecule **a** is −11.7(14)°; this shows that the molecule is twisted, however, to a lesser extent than in the complex [(1,1′-biisoquinoline)$PtCl_2$] and the 3,3′-bisubstituted 2,2′-bipyridine complexes. The calculated rotational barrier required to adopt this conformation (torsion angle N(2)−C(1)−C(19)−N(18) equals −11.7(14)°) has been estimated to be E_{rot} = 6.39 kcal/mol. Neglecting the effects of entropy, $E_{rot} \approx \Delta G_{rot} = -RT\ln K$ (ΔG_{rot} is free enthalpy of rotation), hence, the equilibrium constant, K, for rotation between the conformers can be estimated. Therefore, $K_{rot} \approx 2.06 \times 10^{-5}$, which indicates that there is one molecule of the less stable conformer for every 48500 molecules of the more stable conformer. (For comparison, the rotational barrier E_{rot} for 1,1′-biisoquinoline (from 85.06° to 35.18°) is 4.97 kcal/mol; $K_{rot} = 2.28 \times 10^{-4}$; 1 molecule of the less stable conformer for every 4387 molecules of the more stable conformer.)

The results obtained indicate that the concentration of the proper conformer of L-OCH_3 [torsion angle N(2)−C(1)−C(19)−N(18) equals −11.7(14)°] is sufficient for complexation to occur at standard temperatures. Moreover, protogenic polar solvents can stabilize this conformation by the formation of hydrogen bonds, thus the energy barrier for rotation may decrease and complex formation is made easier. This is supported by the experiment in which [(L-OCH_3)$PtCl_2$] is formed.

Antiproliferative Activity

The in vitro growth-inhibitory effect of the ligands L-OH, L-OAc, L-OMe were tested on four human cancer cell lines: A549, SW707, HCV29T, T47D and on mice leukemia L1210. The results, expressed as the IC_{50} values, are presented in Table 7. In general, the ligands are much more

Table 7. The in vitro antiproliferative activity against human cancer cell lines and mice leukemia

Cell line: Compound	A549 IC_{50} [μmol/dm³]	SW707	HCV29T	T47D	L1210
L-OH	9.56	10.19	9.25	9.79	1.7
L-OAc	18.97	21.24	17.83	11.66	2.3
L-OCH_3	neg.	25.04	20.55	17.47	−
[Pt(L-OH)Cl_2][a]	−	−	−	−	ca. 10
[Pt(L-OAc)Cl_2][a]	−	−	−	−	ca. 10
Carboplatin	108.00	210.00	68.70	202.00[b]	−
Adriamycin	−	−	−	−	0.03
Elliptinium acetate	−	−	−	−	0.07

[a] Slurry in DMSO. [b] IC_{30}; neg. inactive; − not measured.

active against the human lines than the reference carboplatin. The most active was L-OH (ID_{50} against all human cancer cell lines and against L1210). The least active was L-OCH_3 (negative against A549).

The relatively insoluble platinum complexes, [(L-OH)$PtCl_2$] and [(L-OAc)$PtCl_2$], were tested against L1210; they could be measured only as a slurry in DMSO. Although IC_{50} values for the complexes are less than those for the reference compounds, as well as for the ligands, their activity seems to be significant, taking into account that the cytotoxic effect was only caused by a soluble part of sample. More advanced studies will be described elsewhere.[15]

The results obtained indicate that the new olivacine derivatives and their neutral platinum complexes are promising cytotoxic agents of a new class; however, improvement on the solubility of the chloro complexes is necessary. This may be achieved by exchanging the chloride ligands for chosen dicarboxylate ligands, according to our earlier methodology.[57]

Experimental Section

Materials: The starting material for the synthesis of the new 9-olivacine derivatives was 2-(6-methoxy-1-methylcarbazol-2-yl)-ethylamine, which was synthesized from 5-methoxyindol and 4-acetylpyridine, both purchased from Aldrich.

The starting material for the synthesis of the platinum complexes were the new 9-olivacine derivatives, and K_2PtCl_4, which was prepared from metallic Pt by known procedure.[58] Synthetic procedures for the platinum complexes were carried out in the dark. All other chemicals obtained from commercial suppliers were used as received and were of analytical grade.

Physical Measurements: Elemental analyses (C,H,N) were performed on a model Perkin−Elmer Elemental Analyzer 2400 CHN. Pt was analyzed by ICP-AES method on ARL 3410 instrument (with permissible deviation ± 5%), with previous mineralization by microwaves on MDS 200 CEM mineralizator. Thin-layer chromatography was carried out on pre-coated silica gel plates (Poligram) and on plates with the UV marker (Silufol, Cavalier). (A) 1-propanol-1:acetic acid:water (4:16:1) and (B) acetone/chloroform (1:4) were used as developing mixtures. The spots were visualized with rubeanic acid (VEB Laborchemie Apolda) (10% in acetone) and with UV radiation (254 and 366 nm).

Conductivity data (Λ_0 = immediate, Λ_{24} = after twenty four hours [$\Omega^{-1}\cdot mol^{-1}\cdot cm^2$]) were obtained at 298 K in DMF solutions (c = 4.7×10^{-5} M), on a Radelkis OK-104 conductometer.

IR and FIR spectra were run as KBr pellets and nujol mulls, respectively, on a Bruker JFS 113 V FT-IR spectrometer. UV spectra were obtained on Hewlett Packard HP8452A diode array spectrophotometer. ESI-MS measurements were done on a Finnigan MAT TSQ-700 triple-quadrupole mass spectrometer (equipped with a 20 kV conversion dynode for enhanced sensitivity). The ion signals were recorded by a Finnigan ICIS data system operated on a DEC station 3000. The ^1H spectra were recorded at 500.13 MHz and ^{13}C NMR spectra at 125.72 MHz on a Bruker Avance 500 instrument (all at 298 K). Spectra were referenced to the internal solvent peaks.

X-ray Structure Determination: X-ray diffraction data were collected at 100 K on a KUMA KM4CCD diffractometer with graphite−monochromated Mo-K_α radiation (λ = 0.71073 Å) using the ω scan technique. The crystal structures were solved with SHELXS-97 and refined against $|F|^2$ using SHELXL-97.[59]

In the ligands L-OCH_3 (**5**) and L-OAc (**7**), all hydrogen atoms were found from the difference maps and refined isotropically. In the complex [(L-OCH_3)$PtCl_2$] (**8**) all hydrogen atoms were placed in their geometrically calculated positions d(C−H) = 1.08 Å. Platinum atoms and chloride ligands were refined anisotropically. One of the DMF molecules is disordered, with an occupation factor of 0.5. Refinement of the water molecule gave an occupation factor of 0.5.

Single crystals of the ligands were obtained at ambient temperature by slow evaporation of solvents. L-OCH_3 (**5**) was crystallized from dichloromethane and L-OAc (**7**) from methanol. The nucleation process of [(L-OCH_3)$PtCl_2$] (**8**) crystallization was started after dropwise addition of diethyl ether to the DMF solution of **8**. CCDC-203648, -203649, and -203650 contain the supplementary crystallographic data for this paper. These data can be obtained free of charge at www.ccdc.cam.ac.uk/conts/ retrieving.html [or from the Cambridge Crystallographic Data Centre, 12 Union Road, Cambridge CB2 1EZ, UK; Fax: (internat.) + 44-1223-336-033; E-mail: deposit@ccdc.cam.ac.uk].

Computational Methods: Quantum mechanical calculations were performed using semiempirical PM3 methods[60][61] with the computer software − HyperChem. Pro ver. 7.[62] SCF and geometry optimization − Polak−Ribiere (conjugate gradient) minimization algorithm, SCF convergence limit was set to 10^{-6}, energy optimization gradient was set to 0.01 kcal/(mol × Å).

In-Vitro Cytotoxicity Assays: Test on human cancer cell lines (A549, SW707, HCV29T, T47D) were performed at the Institute of Immunology and Experimental Therapy, Polish Academy of Sciences, in Wroclaw, Poland, as described.[15] The test on mice leukemia L1210 was performed at the Curie Institute − Orsay (France), as described.[15]

2-(6-Methoxy-1-methylcarbazol-2-yl)ethylamine (1): Compound **1** was obtained in accordance with ref.[12]

***N*-[2-(6-Methoxy-1-methyl-9*H*-carbazol-2-yl)ethyl]-5-methyl-2-pyridinecarboxamide (2):** Triethylamine (506 mg, 5.0 mmol) was added to the 5-methyl-2-pyridinecarboxylic acid (603 mg, 4.4 mmol) in dry dichloromethane (100 mL). To the resulting mixture, after cooling to 263 K, a solution of ethyl chloroformate (543 mg, 5.0 mmol) in dry dichloromethane (10 mL) was added, whilst stirring. This was stirred for a further 30 min and then 2-(6-methoxy-1-methylcarbazol-2-yl)ethylamine (**1**) (1017 mg, 4.0 mmol) in THF (100 mL) was added dropwise at 263 K, and left to reach room temperature (20 h) whilst stirring. The precipitate was collected and the filtrate was evaporated to dryness. The residue dissolved in water (50 mL), was made basic with concentrated aqueous ammonia and extracted with dichloromethane and dried over magnesium sulfate. Evaporation of the solvent provided a solid residue, which was recrystallized from ethanol to give 900 mg, (60%) of amide **2**. M.p. 458 K. $C_{23}H_{23}N_3O_2$ (373.5): calcd. C 73.97, H 6.21, N 11.25; found C 73.56, H 6.54, N 10.91. ^1H NMR ($CDCl_3$, 298 K): δ = 2.36 (s, 3 H, 5′-CH_3), 2.52 (s, 3 H, 1-CH_3), 3.12 [t, J(α-CH_2−β-CH_2) = 6.2 Hz, 2 H, α-CH_2], 3.70 (t, 2 H, β-CH_2), 3.90 (s, 3 H, 6-OCH_3), 7.05−7.11 (m, 2 H, 3-H +7-H), 7.40 (d, $J_{8,7}$ = 8.7 Hz, 1 H, 8-H), 7.50 (d, $J_{5,7}$ = 2.5 Hz, 1 H, 5-H), 7.74 (dd, $J_{4',3'}$ = 8 Hz, 1 H, 4′-H), 7.84 (d, $J_{4,3}$ = 7.8 Hz, 1 H, 4-H),

8.03 (d, $J_{3',4'}$ = 8 Hz, 1 H, 3′-H), 8.31 (s, 1 H, 6′-H), 8.75 (s, 1 H, 9-H).

9-Methoxy-5-methyl-1-(5′-methylpyridin-2-yl)-3,4-dihydro-6*H*-pyrido[4,3-*b*]carbazole (3): The preceding amide (**2**) (1867 mg, 5.0 mmol) was dissolved in boiling toluene (150 mL) and treated dropwise with phosphorous oxychloride (12 mL). Reflux was continued for a 12 h, and evaporation under reduced pressure afforded a residue, which was dissolved in water (100 mL), was made basic to pH 9−10 with concentrated aqueous ammonia, and extracted with dichloromethane. Evaporation of the solvent provided a solid residue, which was recrystallized from ethanol to give yellow crystals (1320 mg, 74%). M.p. 554 K. $C_{23}H_{21}N_3O$ (355.4): calcd. C 77.72, H 5.96, N 11.82; found C 77.52, H 6.03, N 11.91. ^{1}H NMR ($CDCl_3$, 298 K): δ = 2.45 (s, 3 H, 5′-CH_3), 2.68 (s, 3 H, 5-CH_3), 2.83 (t, $J_{4,3}$ = 6.5 Hz, 2 H, 4-H), 3.85 (s, 3 H, 6-OCH_3), 3.90 (t, $J_{3,4}$ = 6.5 Hz, 2 H, 3-H), 7.00 (dd, $J_{8,7}$ = 8.7, $J_{8,10}$ = 2.4 Hz, 1 H, 8-H), 7.34 (d, $J_{7,8}$ = 8.8 Hz, 1 H, 7-H), 7.41 (d, $J_{10,8}$ = 2.3 Hz, 1 H, 10-H), 7.52 (dd, $J_{4',3'}$ = 8.5 Hz, 1 H, 4′-H), 7.88 (d, $J_{3',4'}$ = 8.6 Hz, 1 H, 3′-H), 8.38 (br. s, 1 H, 6-H), 8.70 (s, 1 H, 6′-H), 9.07 (s, 1 H, 11-H).

9-Methoxy-5-methyl-1-(5′-methylpyridin-2-yl)-6*H*-pyrido[4,3-*b*]carbazole (4): Compound **3** (1422 mg, 4.0 mmol) was refluxed in diphenyl ether (50 mL) in the presence of 10% palladized charcoal (100 mg) for 30 min. After removal of the catalyst, the filtrate was cooled and diluted with hexane. The resulting precipitate was collected and washed with hexane and recrystallized from ethyl acetate to give yellow crystals (1060 mg, 75%). M.p. 544 K. $C_{23}H_{19}N_3O$ (353.4): calcd. C 78.17, H 5.42, N 11.89; found C 77.97, H 5.66, N 11.73. ^{1}H NMR ($CDCl_3$, 298 K): δ = 2.34 (s, 3 H, 5′-CH_3), 2.56 (s, 3 H, 5-CH_3), 3.85 (s, 3 H, 9-OCH_3), 7.12 (dd, $J_{8,7}$ = 8.8, $J_{8,10}$ = 2.4 Hz, 1 H, 8-H), 7.26 (d, $J_{7,8}$ = 8.7 Hz, 1 H, 7-H), 7.65 (d, $J_{10,8}$ = 2.4 Hz, 1 H, 10-H), 7.87 (dd, $J_{4',3'}$ = 8.4 Hz, 1 H, 4′-H), 7.97 (m, 2 H, 4H +3′-H), 8.49 (d, $J_{3,4}$ = 6.5 Hz, 1 H, 3-H), 8.55 (s, 1 H, 6-H), 8.81 (s, 1 H, 6′-H), 9.12 (s, 1 H, 11-H).

9-Methoxy-5,6-dimethyl-1-(5′-methylpyridin-2-yl)-6*H*-pyrido[4,3-*b*]carbazole (5, L-OCH_3): A mixture of **4** (1060 mg, 3.0 mmol), fine powdered dry potassium carbonate (750 mg), dimethyl carbonate (20 mL), dimethyl formamide (3 mL) and 18-crown-6-ether (3 drops) was heated under reflux, whilst stirring for a 12 h. After evaporation to dryness, the residue was dissolved in water. The solid was collected, air-dried, and recrystallized from isopropyl acetate to give yellow crystals (880 mg, 80%). M.p. 497−498 K. R_f = 0.82 (A), 0.34 (B). $C_{24}H_{21}N_3O$ (367.45): calcd. C 78.45, H 5.76, N 11.44; found C 77.09, H 5.93, N 11.50. MS: *m*/*z* (%) = 368 (100).

9-Hydroxy-5,6-dimethyl-1-(5′-methylpyridin-2-yl)-6*H*-pyrido[4,3-*b*]carbazole (6, L-OH): A mixture of (**L-OCH_3**) (735 mg, 2 mmol) and hydrobromic acid (48%, 30 mL) was heated under reflux, whilst stirring for 3 h. After evaporation to dryness, the residue was dissolved in water. The resulting mixture was made basic with concentrated ammonia and extracted with dichloromethane. The extract was dried over magnesium sulfate and the solvents evaporated. The solid was recrystallized from dichloromethane to give yellow crystals (676 mg, 96%). M.p. 522−524 K. R_f = 0.82 (A). $C_{23}H_{19}N_3O$ (353.42): calcd. C 78.17, H 5.42, N 11.89; found C 78.01, H 5.61, N 11.72. MS: *m*/*z* (%) = 354 (100).

9-Acetoxy-5,6-dimethyl-1-(5′-methylpyridin-2-yl)-6*H*-pyrido[4,3-*b*]carbazole (7, L-OAc): A solution of (**L-OH**) (106 mg, 0.3 mmol) in dry pyridine (1 mL) was cooled to 263 K and then treated dropwise with acetic anhydride (1 mL). The mixture was stirred at 263 K for 30 min and allowed to reach room temperature over 3 h. The mixture was then poured into cold water (10 mL) and extracted with dichloromethane. Evaporation of the solvent provided a solid residue, which was recrystallized from isopropyl acetate to give yellow crystals (105 mg, 89%). M.p. 529−530 K. R_f = 0.80 (A); 0.31 (B). $C_{25}H_{21}N_3O_2$ (395.46): calcd. C 75.93, H 5.35, N 10.62; found C 75.18, H 5.41, N 10.43. MS: *m*/*z* (%) = 396 (100).

Dichloro[(9-methoxy-5,6-dimethyl-1-(5′-methyl-pyridin-2-yl)-6*H*-pyrido[4,3-*b*]carbazole]platinum(II) [8, (L-OCH_3)$PtCl_2$]: 9-Methoxy-5,6-dimethyl-1-(5-methylpyridin-2-yl)-6*H*-pyrido[4,3-*b*]carbazole (**L-OCH_3**) (150 mg, 0.41 mmol) was dissolved in methanol (250 mL) and an aqueous solution (8 mL) of K_2PtCl_4 (169 mg, 0.41 mmol) was added dropwise. The mixture was stirred at room temperature for several hours and left for 5 weeks. The precipitate obtained was centrifuged, washed with a small amount of water to remove KCl (tested with $AgNO_3$). The platinum complex was dried over P_4O_{10} in vacuo. Yield 239 mg (92%). R_f = 0.88 (A), 0.76 (B). $C_{24}H_{21}Cl_2N_3OPt$ (633.44): calcd. C 45.51, H 3.34, Cl 11.19, N 6.63, Pt 30.68; found C 44.77, H 3.29, N 6.42, Cl 11.08, Pt 30.68. MS: *m*/*z* (%) = 633 (100). $\Lambda_{0\ and\ 24}$ = 4.2 [$\Omega^{-1}\cdot mol^{-1}\cdot cm^2$].

Dichloro[(9-hydroxy-5,6-dimethyl-1-(5′-methylpyridin-2-yl)-6*H*-pyrido[4,3-*b*]carbazole]platinum(II) [9, (L-OH)$PtCl_2$]: 9-hydroxy-5,6-dimethyl-1-(5′-methylpyridin-2-yl)-6*H*-pyrido[4,3-*b*]carbazole (**L-OH**) (149 mg, 0.42 mmol) was dissolved in methanol (200 mL), an aqueous solution (8 mL) of K_2PtCl_4 (175 mg, 0.42 mmol) was then added. The mixture was stirred at room temperature for several hours and left for 4 weeks. The precipitate obtained was centrifuged, washed with a small amount of water to remove KCl (tested with $AgNO_3$) and dried over P_4O_{10} in vacuo. Yield 221 mg (85%). R_f = 0.94 (A). $C_{23}H_{19}Cl_2N_3OPt$ (619.41): calcd. C 44.6, H 3.09, Cl 11.45, N 6.78, Pt 31.49; C 44.33, H 3.68, Cl 10.68, N 6.46, Pt 28.44. MS: *m*/*z* (%) = 620 (100). $\Lambda_{0\ and\ 24}$ = 5.0 [$\Omega^{-1}\cdot mol^{-1}\cdot cm^2$].

Dichloro[9-acetoxy-5,6-dimethyl-1-(5′-methyl-pyridin-2-yl)-6*H*-pyrido[4,3-*b*]carbazole]platinum(II) [10, (L-OAc)$PtCl_2$]: 9-Acetoxy-5,6-dimethyl-1-(5-methylpyridin-2-yl)-6*H*-pyrido[4,3-*b*]carbazol] (**L-OAc**) (140 mg, 0.35 mmol) was dissolved in methanol (150 mL), an aqueous solution (9 mL) of K_2PtCl_4 (145 mg, 0.35 mmol) was then added. The mixture was stirred at room temperature for several hours and then left for about 4 weeks. The precipitate obtained was centrifuged, washed with a small amount of water to remove KCl (tested with $AgNO_3$). The platinum complex was dried over P_4O_{10} in vacuo. Yield 190 mg (82%). R_f = 0.72 (B). $C_{25}H_{21}Cl_2N_3O_2Pt$ (661.45): calcd. C 45.40, H 3.20, Cl 10.72, N 6.35, Pt 29.49; found C 44.58, H 3.74, Cl 10.05, N 6.00, Pt 25.49. MS: *m*/*z* (%) = 663 (100). $\Lambda_{0\ and\ 24}$ = 3.3 [$\Omega^{-1}\cdot mol^{-1}\cdot cm^2$].

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