Non-Platinum-Group Metal Antitumor Agents: History, Current Status, and Perspectives

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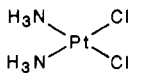
Contents

I. Introduction

The scope of this paper is limited to cytostatically active complexes including at least one metal atom. In principle, three different groups of metal-containing antitumor agents can be distinguished: inorganic complexes composed of a central metal atom surrounded by inorganic ligands [The most famous example of inorganic cytostatics is *cis*-diamminedichloroplatinum(II) (cisplatin).]; organometallic complexes containing one or more metal atoms as well as organic ligands, the ligands being linked to the metals by direct carbon–metal bonds [Many of the non-platinum-group metal antitumor agents known are typical organometallic compounds.]; complexes also including metal atoms and organic ligands but lacking carbon–metal bonds, therefore not defined as organometallic [Interestingly, many of the antitumor platinum complexes of the second and third generation as well as some non-platinum-group metal cytostatics belong to this group of compounds.].

II. Platinum and Platinum-Group Metal Antitumor Agents

The recent history of metal-containing antitumor agents began with the detection of antitumor properties for the inorganic compound *cis*-diamminedichloroplatinum(II) (cisplatin, **1**) in 1969.[1] In the meantime,



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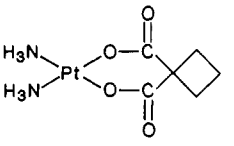
the considerable clinical success of this inorganic cytostatic drug against numerous human malignancies, especially against urogenital tumors and carcinomas of the head and neck, initiated a broad search for further antitumor platinum compounds. Numerous platinum complexes of the second generation have been developed; about 10 of them entered clinical studies in the late 1970s.[2] The main representatives of second-generation platinum complexes are diammine(cyclobutane-1,1-dicarboxylato)platinum(II) (carboplatin, **2**) and aquo[1,1-bis(aminomethyl)cyclohexane](sulfato)-platinum(II) (spiroplatin, **3**), which like cisplatin are four-coordinate, planar complexes, and bis(isopropylamine)-*cis*-dichloro-*trans*-dihydroxoplatinum(IV) (iproplatin, **4**), which in contrast contains platinum(IV) in an octahedral coordination sphere. These platinum complexes, which contain organic ligands but lack carbon–metal bonds, are in comparison to **1** characterized by a similar clinical spectrum of activity, by reduced nephrotoxicity, and obviously by increased myelotoxicity.[3,4]



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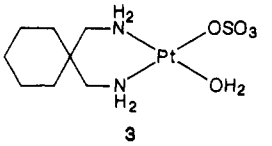

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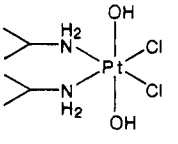

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Not only platinum complexes exhibit antitumor properties; numerous complexes containing platinum-group metals such as ruthenium, rhodium, or palladium are characterized by cytostatic activity. This was shown for octahedral, binuclear rhodium(II) carboxylates[5] such as rhodium(II) propionate (**5**, R = CH_2CH_3) and rhodium(II) butyrate (**5**, R = $CH_2CH_2CH_3$), octahedral ruthenium(II) and ruthenium(III) complexes[6] such as *cis*-$[(NH_3)_4RuCl_2]Cl$ (**6**) and *cis*-$[(dmso)_4RuCl_2]$ (dmso = dimethyl sulfoxide), and planar and octahedral palladium(II) and palladium(IV) complexes[7] such as *cis*-

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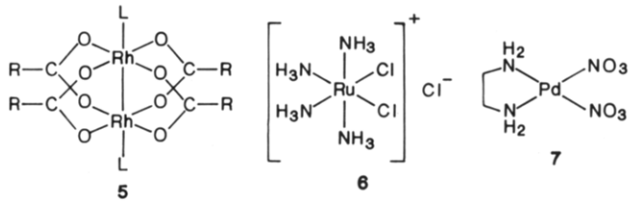
Professor Dr. Hartmut Köpf was born in 1937 in Esslingen, Germany, began his chemical education at the University of Munich, and received his Ph.D. in 1963 under the supervision of Professor Dr. Max Schmidt at the University of Marburg. In the following years, he performed research work on titanocene pentasulfide and related complexes in the group of Professor Schmidt at the Universities in Marburg and Würzburg. 1975, he was appointed Professor of Inorganic Chemistry at the Technical University of Berlin. His current research interests are still concerned with metallocene complexes, e.g. dithiolene and dichalkogenolene chelates, of the early transition metals and with the antitumor properties of such species. Professor Köpf's work is published in about 100 papers.

Professor Dr. Petra Köpf-Maier was born in 1952 in Hof, Bavaria, Germany. She studied medicine in Würzburg and Berlin, became a physician in 1977, and received her M.D. degree from the Free University of Berlin in 1978. Professor Köpf-Maier was appointed Professor of Anatomy at the University of Ulm in 1984, at the Free University of Berlin in October 1986. She is the author of over 70 publications. Her current research interests involve the biological properties of platinum and non-platinum-group metal antitumor agents, especially of metallocene and metallocenium compounds. She is performing antitumor testing in vivo and in vitro, toxicologic investigations, and morphologic and cytokinetic studies into the influence of inorganic and organometallic antitumor agents upon experimental and human tumors.

$[(en)Pd(NO_3)_2]$ (en = 1,2-diaminoethane) (**7**) and *cis*-$[(NH_3)_2PdCl_4]$. Interestingly, against some experimental tumor systems, rhodium complexes effected equal antitumor activity as **1**, their toxic features being more favourable than those of **1**.[8]

III. Non-Platinum-Group Metal Antitumor Agents

Regarding non-platinum-group metal cytostatics, main-group metal compounds can be distinguished from transition-metal complexes.



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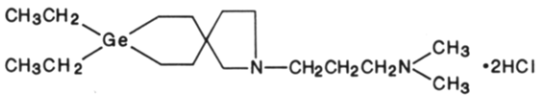
A. Main-Group Metal Compounds

Gallium salts[9] were the first main-group compounds for which antiproliferative activity was described (in 1975). Other important main-group cytostatics contain metals neighboring gallium, i.e. germanium and tin.

Antitumor gallium compounds are inorganic salts regarded to be composed of Ga^{3+} cations and anions of different kind. The most effective gallium compound is gallium(III) nitrate (**8**).[9] Its antiproliferative activity was established against Walker carcinosarcoma and various experimental sarcomas. Clinical studies, which have been performed since 1976 against numerous human tumors, only revealed limited activity against Hodgkin's and non-Hodgkin's lymphomas.[4] The pattern of side effects of **8** is similar to that of **1**, renal and gastrointestinal toxicities being the main and dose-limiting side effects.[4,10]

Within main-group IV (14),[55] germanium compounds were the first ones for which antiproliferative properties were detected. Presently, there are two main representatives of antitumor germanium compounds: the monomeric 8,8-diethyl-2-[3-(*N*-dimethylamino)-propyl]-2-aza-8-germaspiro[4.5]decane (spirogermanium, **9**)[11] and the polymeric bis[(carboxyethyl)germanium] trioxide (germanium sesquioxide, Ge-132, **10**).[12] In contrast to gallium salts, both **9** and



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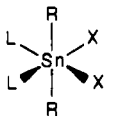
10 are typical organometallic complexes containing carbon–metal bonds. Their antitumor activities have been established against various experimental tumors such as Ehrlich ascites tumor and Lewis lung carcinoma. In clinical studies, limited value was revealed for antitumor germanium complexes. In the case of **9**, tumor-inhibiting effects were observed against advanced ovarian carcinomas and lymphocytic lymphoma.[4] Interestingly, the antitumor activity of organogermanium compounds is apparently not based on direct cytotoxic effects, but on host-mediated, immunopotentiating mechanisms.[13] Especially, activation of interferon seems to be an important factor. This was confirmed for **9** as well as for **10**. Therefore, both compounds are currently undergoing clinical studies as biological response modifiers.

The third group of antitumor main-group metal complexes detected so far is represented by diorganotin(IV) derivatives such as the octahedral diorganotin dihalide complexes $R_2SnX_2L_2$ [**11**: R = alkyl, phenyl; X = F, Cl, Br, I, NCS; L = unidentate ligand, (e.g. pyridine); L_2 = bidentate chelating ligand (e.g. 2,2′-



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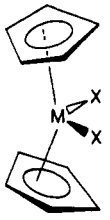
bipyridyl or *o*-phenanthroline)] and di- and triorganotin complexes of nitrogenous bases and amino acids, e.g. R_2Sn[adeninate(1−)]$_2$ and R_2Sn[L-cysteinate(2−)].[14,15] Their antiproliferative activity was detected in 1980 and later against P388 leukemia.[14] Though this activity is strong and comparable to that of **1**, further studies showed that there is obviously no activity against other experimental tumor systems, for example against leukemia L1210, B16 melanoma, colon 38 carcinoma, and heterotransplanted human tumors.[16] Nevertheless, it is worth considering that antitumor tin complexes structurally resemble **1** or other cytostatic platinum-metal complexes with respect to the presence of a *cis*-dihalometal moiety.

B. Transition-Metal Compounds

Antitumor compounds of transition metals other than platinum-group metals either contain early transition metals like titanium and vanadium, medium transition metals like iron, or late transition metals like copper and gold. *Early*-transition-metal compounds showing tumor-inhibiting efficacy are mainly represented by metallocene complexes of type **12** (M = Ti, V, Nb, Mo;



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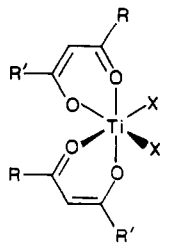
e.g., X = Cl).[17] These are organometallic complexes, the cyclopentadienyl ring ligands being bound to the central metal atom M by carbon–metal bonds. Metallocene complexes **12** have shown antiproliferative activity against various experimental tumors, e.g. Ehrlich ascites tumor, sarcoma 180, B16 melanoma, colon 38 carcinoma, and Lewis lung carcinoma, as well as against xenografted human carcinomas.[17,18]

Other antitumor titanium complexes are the six-coordinate bis(benzoylacetonato)titanium(IV) dihalides and bis(alkoxides) **13** (R = CH_3, R′ = C_6H_5, X = F, Cl, Br, OC_2H_5). Their antitumor activity has been established against Walker 256 carcinosarcoma, leukemia L1210, and sarcoma 180;[19] some similar zirconium and hafnium complexes have been found to be active against sarcoma 180.[20]



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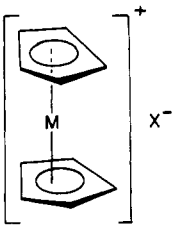
Whereas the antitumor metallocene dihalides **12** are neutral compounds, the cytostatically active *medium*-transition-metal agents are saltlike metallocenium complexes **14** (M = Fe, Co; e.g., $X^- = FeCl_4^-$) containing in the cation the coordinatively saturated central metal atom interposed between two parallel cyclopentadienyl ring ligands. Thus, metallocenium compounds are also organometallic complexes. Their antitumor activity was detected in 1984 against Ehrlich ascites tumor.[21]



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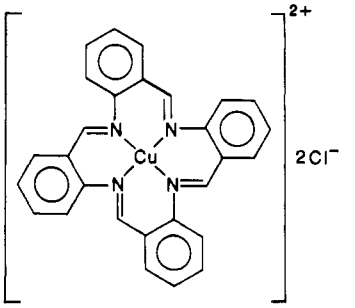
Regarding *late*-transition-metal antitumor agents, numerous compounds of copper are known that contain organic ligands exhibiting antiproliferative activity by themselves. Prominent examples of this type of cytostatic compounds are bleomycin- and thiosemicarbazone–copper complexes.[22] Besides these, many other copper complexes containing, e.g. ligands of the Schiff base type, have shown antiproliferative activity. Thus, efficacy against experimental animal tumors was revealed for the neutral complex *trans*-bis(salicylaldoximato)copper(II)[23] as well as for the copper(II) complex salt **15** of the macrocyclic ligand tetrabenzo[*b*,*f*,*j*,*n*]-1,5,9,13-tetraazacyclohexadecene.[24]



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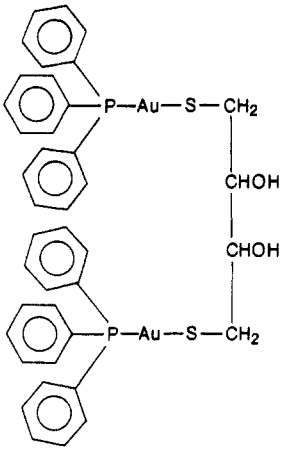
From certain gold(I) complexes like auranofin it is known that they are clinically well-approved therapeutic drugs against rheumatoid arthritis.[25] Reports upon in vivo antitumor activity of gold complexes, however, are comparably rare.[26] Complexes for which in vivo antitumor activity has been described recently are gold(I)–phosphine complexes such as the neutral binuclear **16** containing a bridging dithiolate ligand[27] and the saltlike, mononuclear, tetrahedral chelate complex **17** comprised of two bidentate 1,2-bis(diphenylphosphino)ethane ligands.[28] They effected antiproliferative effectivity against Ehrlich ascites tumor or, respectively, against P388 leukemia, M5076 reticulum cell sarcoma, and mammary adenocarcinoma 16/c.

IV. Antitumor Properties of Metallocene and Metallocenium Complexes

The following chapter focuses on the current status of development of metallocene and metallocenium complexes as examples of non-platinum-group metal cytostatics.



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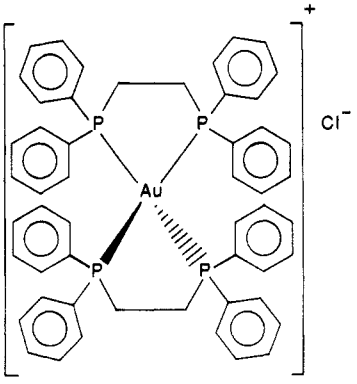

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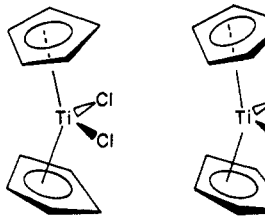
Antitumor early- and medium-transition-metal compounds are mainly represented by titanocene and ferrocenium complexes that fundamentally differ from cytostatic platinum metal complexes. Their antitumor activities were detected in 1979[17] and 1984,[21] respectively. In the meantime, it was possible to confirm the antiproliferative properties of both groups of compounds against a series of fluid and solid experimental tumors as well as against human malignancies.[18]

A. Metallocene Complexes against Experimental Animal Tumors

Metallocene complexes **12** are characterized by the following structural features: They are organometallic, neutral complexes of distorted-tetrahedral coordination geometry. Their organic ligands are represented by two η^5-cyclopentadienyl ring ligands in tilted arrangement. They contain early transition metals of subgroups IV, V, or VI (4–6).[55] Strong antitumor activity was shown for metallocene complexes containing Ti, V, Nb, or Mo as central atoms M. The two acido ligands X are arranged in adjacent, cis-like positions.

The main representatives of antitumor metallocene complexes are given by titanocene dihalides such as titanocene dichloride [$(C_5H_5)_2TiCl_2$, **18**][29] and titanocene dibromide [$(C_5H_5)_2TiBr_2$, **19**],[30] titanocene carboxylates such as titanocene bis(hydrogen maleinate) [$(C_5H_5)_2Ti(OCOCH{=}CHCOOH)_2$, **20**][31] and titanocene bis(trichloroacetate) [$(C_5H_5)_2Ti(OCOCCl_3)_2$, **21**],[31] and vanadocene dihalides such as vanadocene dichloride [$(C_5H_5)_2VCl_2$, **22**].[17b,32]

Compounds **18–22** exhibited strong antiproliferative activity against fluid Ehrlich ascites tumor.[29–31] After application of optimum doses, they all induced cure



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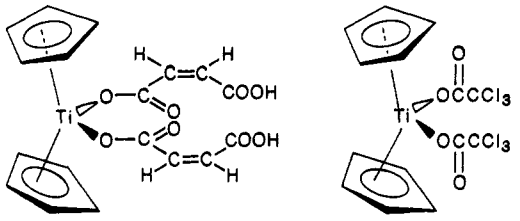

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rates of 100% whereby the values of the therapeutic index, indicating the relation between lethal and effective doses, ranged between 3.3 and 5.5. In Figure 1, the dose–activity and dose–lethality relationships of **18–21** are illustrated after administration to mice bearing Ehrlich ascites tumor. Similar results were obtained against this tumor system with other metallocene dihalides **12** (M = Ti, X = F, I, NCS, N_3;[30] X = Cl, M = Nb, Mo[17c,d]).

In spite of the pronounced antitumor activity against fluid Ehrlich ascites tumor, **18** and **22** showed only marginal activity against the experimental leukemia systems L1210 and P388.[33] This behavior is not limited to metallocene complexes but was also observed with several other promising metal-containing cytostatics, which turned out to be active in test systems other than leukemias.[8,22c,28a] In the case of numerous solid experimental tumors, metallocene complexes were able to clearly suppress tumor proliferation and development. This was shown, for example (Figure 2), for sarcoma 180,[18a] the growth of which was reduced by about 80% after threefold application of 50 mg/kg of **18**, thus that the mean size of treated tumors decreased to 23% of that of control tumors (100%). Similar results were obtained for **19** and **20**.

In the case of solid B16 melanoma (Figure 3), **18** also effected significant and dose-dependent inhibition of tumor growth.[18b] The best T/C values induced by single or threefold doses ranged between 37 and 41%. After application of five doses, the size of treated tumors amounted to only 20% of control tumors. When cisplatin (**1**) was applied in equitoxic doses for comparison purposes, similar values were found. Fivefold injections of 30 or 40 mg/kg of **19** provoked similar tumor inhibition, the T/C ratios amounting to 30 or 26%.

Another animal solid tumor investigated, colon 38 adenocarcinoma, is known from the literature to be inhibited by only few common cytostatics, such as 5-fluorouracil or cyclophosphamide. It was found that **18** (Figure 4) as well as other titanocene derivatives (e.g., **19** and **20**) suppressed tumor proliferation by about 60–80% to 40–20% of control tumor size in a clearly dose-dependent manner.[18b]

Analogous results were obtained for Lewis lung carcinoma.[18d] The growth of this tumor was also significantly reduced by treatment with **18** (Figure 5), **19**, and

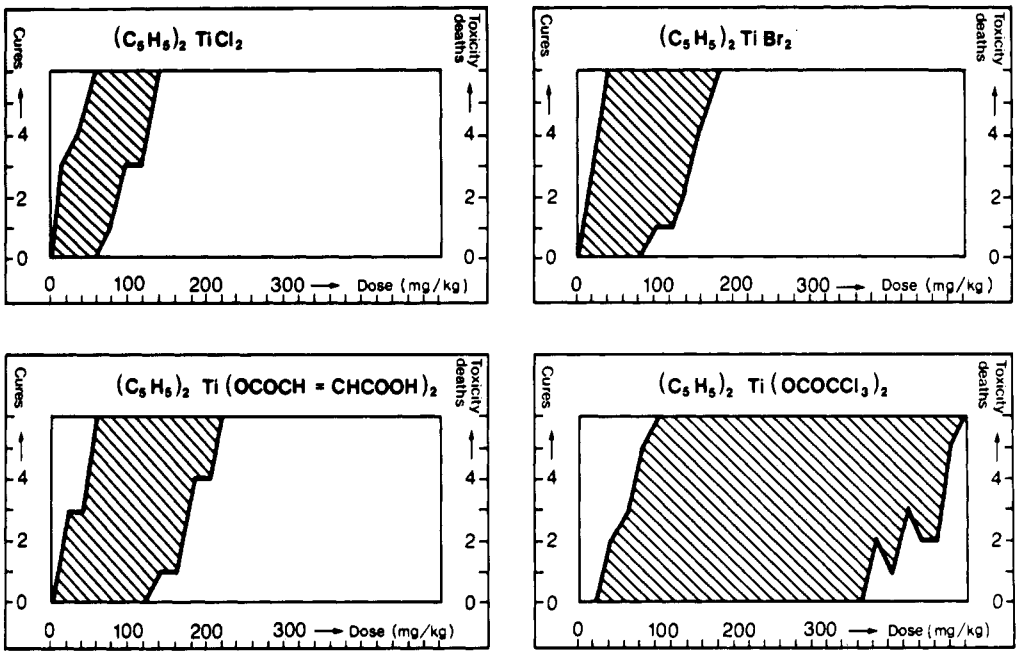


Figure 1. Dose–activity and dose–lethality relationships of titanocene complexes 18–21 against Ehrlich ascites tumor in mice. Cross-hatching indicates surviving animals.

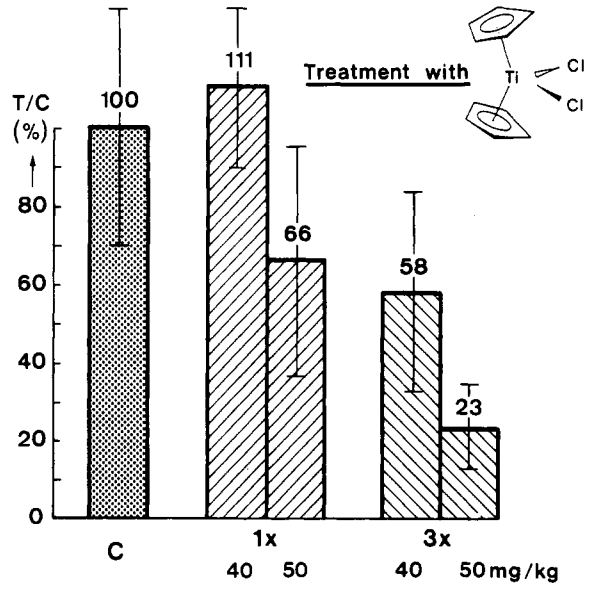


Figure 2. Mean T/C values (tumor weight of treated animals/tumor weight of controls × 100%) determined on day 9 after treatment of mice bearing solid sarcoma 180 with 18 on day 1 (40 or 50 mg/kg, respectively) or on days 1, 3, and 5 (40 or 50 mg/kg) after tumor transplantation.

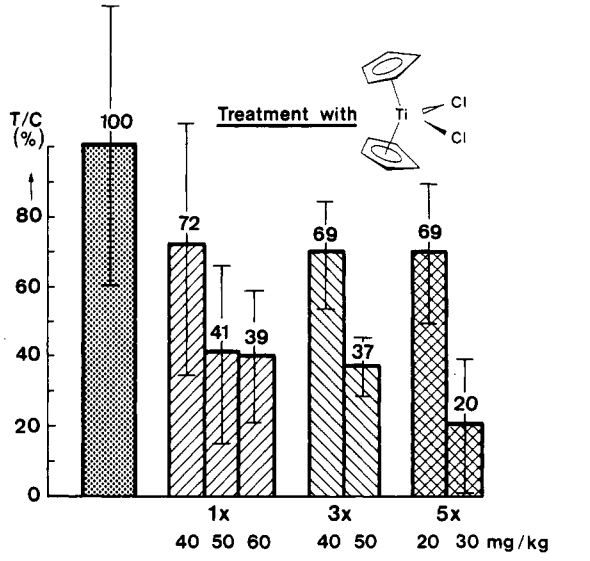


Figure 3. Mean T/C values determined on day 10 after treatment of mice bearing solid B16 melanoma with 18 on day 1 (40, 50, or 60 mg/kg, respectively) or on days 1, 3, and 5 (40 or 50 mg/kg) or determined on day 15 after treatment with 18 on days 1, 3, 5, 7, and 9 (20 or 30 mg/kg).

related compounds. The T/C values that were induced amounted to 70–40%.

In summary of the results of antitumor testing of titanocene complexes against various experimental animal tumors (Figure 6), it becomes evident that $(C_5H_5)_2TiCl_2$ (18) effected good antitumor activity against fluid Ehrlich ascites tumor and sarcoma 180 as well as against a series of solid tumors but that it exhibited only marginal activity against the experimental leukemia systems L1210 and P388. A similar spectrum of activity was recorded for $(C_5H_5)_2TiBr_2$ (19) as well as for the titanocene derivatives 20 and 21, which were modified by introduction of hydrophilic carboxylate ligands as acido ligands X. When the effects caused by the four titanocene complexes 18–21 are compared, slightly superior antitumor activity is detectable for the dichloro derivative 18 in relation to 19–21.

These results underline that the introduction of hydrophilic groups at the position of the acido ligands X within the $(C_5H_5)_2TiX_2$ molecule can be an appropriate method to modify antitumor titanocene complexes and to improve the biological properties of the compounds.[31] Recently, this was confirmed for certain types of ligands other than carboxylato groups.[32] Thus, it is apparently possible to reduce toxic properties, to improve antitumor potency, and to ameliorate water solubility of titanocene complexes.

In this connection it is worth mentioning that the possibility to modify metallocene dihalides at the position of the acido ligands X without loss or affection of the antitumor potency contrasts with the effects re-

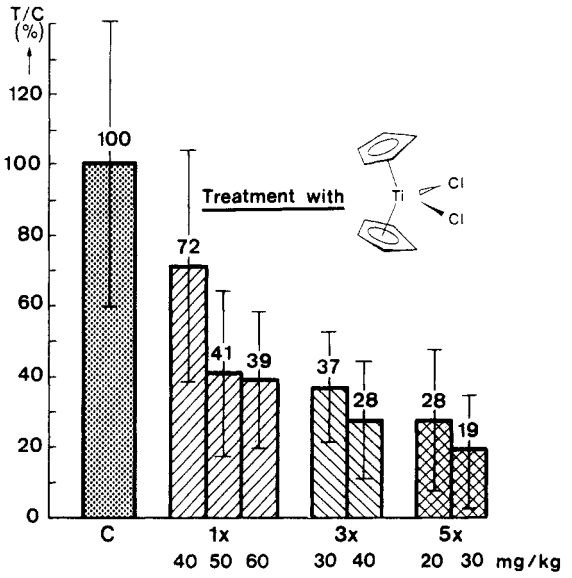


Figure 4. Mean T/C values determined on day 15 after treatment of mice bearing solid colon 38 carcinoma with 18 on day 1 (40, 50, or 60 mg/kg, respectively), on days 1, 3, and 5 (30 or 40 mg/kg), or on days 1, 3, 5, 7, and 9 (20 or 30 mg/kg).

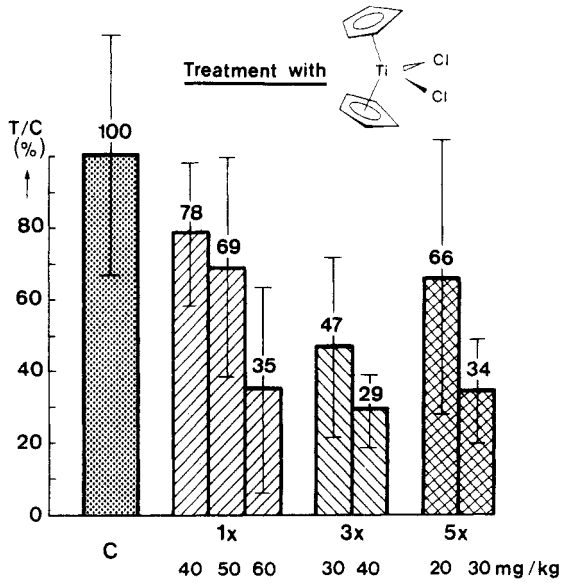
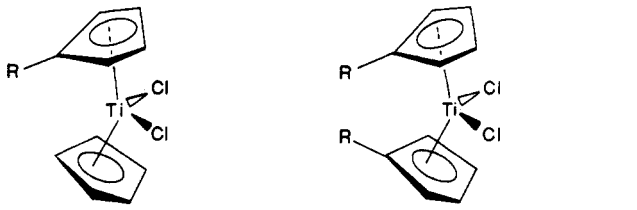
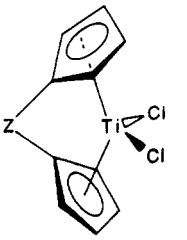


Figure 5. Mean T/C values on day 10 after treatment of mice bearing solid Lewis lung carcinoma with 18 on day 1 (40, 50, or 60 mg/kg, respectively), on days 1, 3, and 5 (30 or 40 mg/kg), or on days 1, 3, 5, 7, and 9 (20 or 30 mg/kg).

sulting from chemical modification at the cyclopentadienyl ring ligands. Substitution of C_5H_5 hydrogen atoms by organic or elemento-organic residues R generally diminishes tumor inhibition, whereby the extent of reduction of antitumor potency is clearly dependent on the degree of modification. This was shown for $(C_5H_5)_2TiCl_2$ (**18**) as a model system against fluid Ehrlich ascites tumor in mice:[35] Monosubstituted titanocene dichloride derivatives **23**, where one hydrogen atom of one cyclopentadienyl ring is formally substituted by a residue R, effected reduced cure rates of 60–80%. 1,1′-Disubstituted (**24**) as well as 1,1′-bridged (**25**) titanocene dichlorides, where both rings are symmetrically modified by introduction of R or connected by a bifunctional group Z, included only sporadic cure rates of 10–30%. Titanocene complexes containing pentamethylated cyclopentadienyl ring ligands, finally,



23: R = C_2H_5, $Si(CH_3)_3$ **24**: R = $Si(CH_3)_3$, $Si(CH_3)_2$-*n*-C_4H_9, $Ge(CH_3)_3$



25: Z = CH_2, $CHCH_3$, $SiHCH_3$, $Si(C_2H_5)_2$, $Ge(CH_3)_2$

were cytostatically inactive against Ehrlich ascites tumor.

This means that the cyclopentadienyl ring ligands apparently represent noli-me-tangere elements and their presence seems to be important for the realization of potent antitumor properties of titanocene complexes.

B. Metallocenium Complexes against Experimental Animal Tumors

Another group of antitumor transition-metal compounds consists of metallocenium complexes **14** showing the following structural characteristics: They are organometallic complexes. They contain two η^5-cyclopentadienyl ring ligands in parallel arrangement. As central metal atom M, they include medium transition metals of subgroup VIII (8)[55] like Fe or Co. Pronounced antitumor activity was shown for ferrocenium compounds. They are ionic, saltlike, and therefore water-soluble complexes composed of metallocenium cations and anions X^-. They do not contain halide ligands covalently bound in cis or cis-like positions and are therefore lacking the *cis*-dihalometal moiety typical for metallocene dihalides (**12**) as well as cisplatin (**1**).

Antitumor ferrocenium complexes $[(C_5H_5)_2Fe]^+X^-$ are represented by compounds with various anions X^-, such as the trichloroacetate **26**[21] with $X^- = CCl_3COO^-$, the picrate **27**[21] with $X^- = 2,4,6\text{-}(NO_2)_3C_6H_2O^-$, the μ-oxo-bis[trichloroferrate(III)] (**28**)[21] with $X^- = {}^1/_2[Cl_3FeOFeCl_3]^{2-}$, or the tetrachloroferrate(III) (**29**)[21] with $X^- = [FeCl_4]^-$. **26–29** showed marked antitumor properties against fluid Ehrlich ascites tumor and induced cure rates of 70–100% over a broad dose range (Figure 7). The appertaining values of the therapeutic index amounted to 1.7–2.0. Other tumors, inhibited by ferrocenium compounds, were solid B16 melanoma, colon 38 carcinoma, or Lewis lung carcinoma.[34] As an example, Figure 8 illustrates tumor inhibition of colon 38 carcinoma by **28**. Tumor growth was clearly suppressed by more than 50 and 70% to T/C ratios of about 30% after application of three- or fivefold injections. Tumor inhibition of similar strength was observed in the case of colon 38 and Lewis lung carcinomas, whereas there was no or only marginal activity against the leukemia systems L1210 and P388. These results confirm that ferrocenium complexes apparently exhibit a spectrum of antitumor activity against ex-

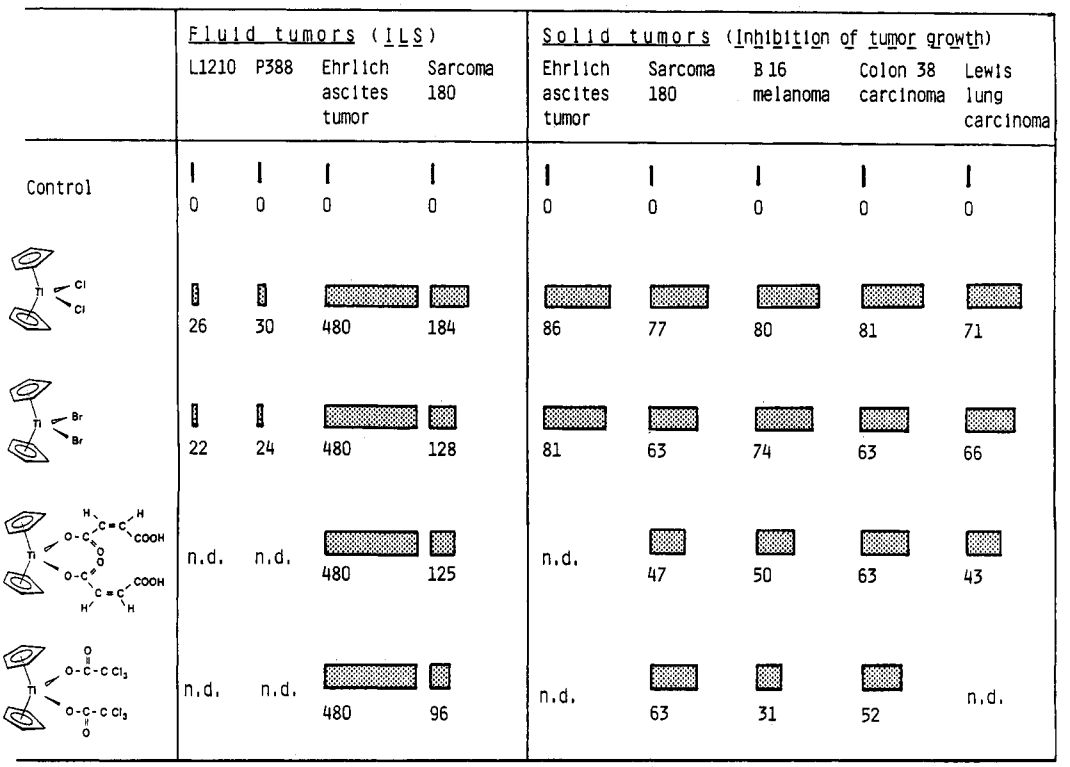


Figure 6. Optimum therapeutic effects of titanocene complexes **18–21** against experimental animal tumors. Parameters evaluated: increase in lifespan (ILS), fluid tumors; inhibition of tumor growth in comparison to untreated controls (100% – T/C), solid tumors. Both parameters expressed as percentages. nd = not determined.

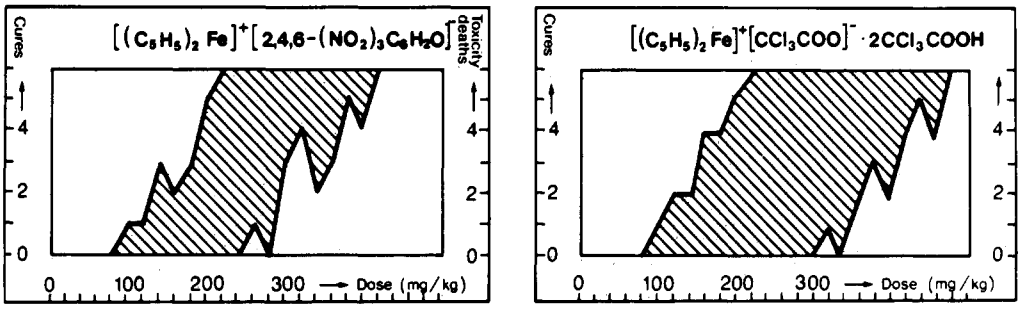


Figure 7. Dose–activity and dose–lethality relationships of ferrocenium complexes **27** and **26** against Ehrlich ascites tumor in mice. Cross-hatching indicates surviving animals.

perimental tumors similar to the case of titanocene complexes.

C. Metallocene and Metallocenium Complexes against Xenografted Human Tumors

Besides antitumor activity against experimental animal tumors, metallocene and ferrocenium complexes also showed antiproliferative efficacy against human tumors xenografted to athymic nude mice. This was verified by pilot experiments performed recently. Diverse human tumors were investigated. Activity was found in the case of human colon adenocarcinomas, human breast carcinoma, human cervix carcinoma and human lung malignancies.

When titanocene complexes **18–20** were administered to animals bearing a human adenocarcinoma derived from the colon sigmoideum, strong tumor inhibition by 70% to T/C ratios of about 30% were effected.[18c] These growth suppressions were highly significant and lasted beyond the end of treatment period (Figure 9). No revival of tumor proliferation took place during at least 3 weeks after the end of treatment. For com-

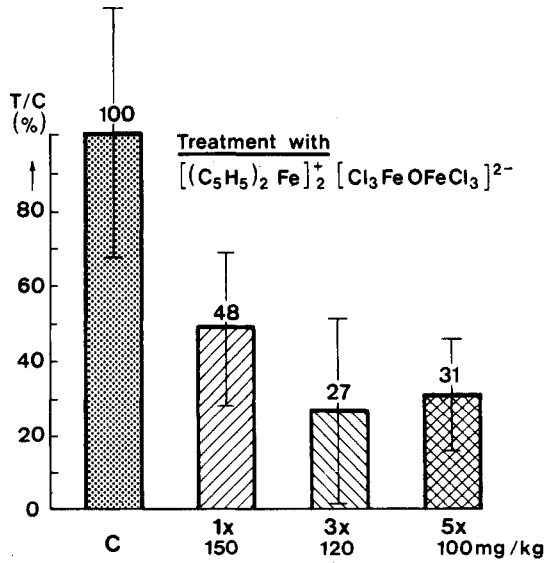


Figure 8. Mean T/C values on day 15 after treatment of mice bearing solid colon 38 carcinoma with **28** on day 1 (150 mg/kg), on days 1, 3, and 5 (120 mg/kg), or on days 1, 3, 5, 7, and 9 (100 mg/kg) after tumor transplantation.

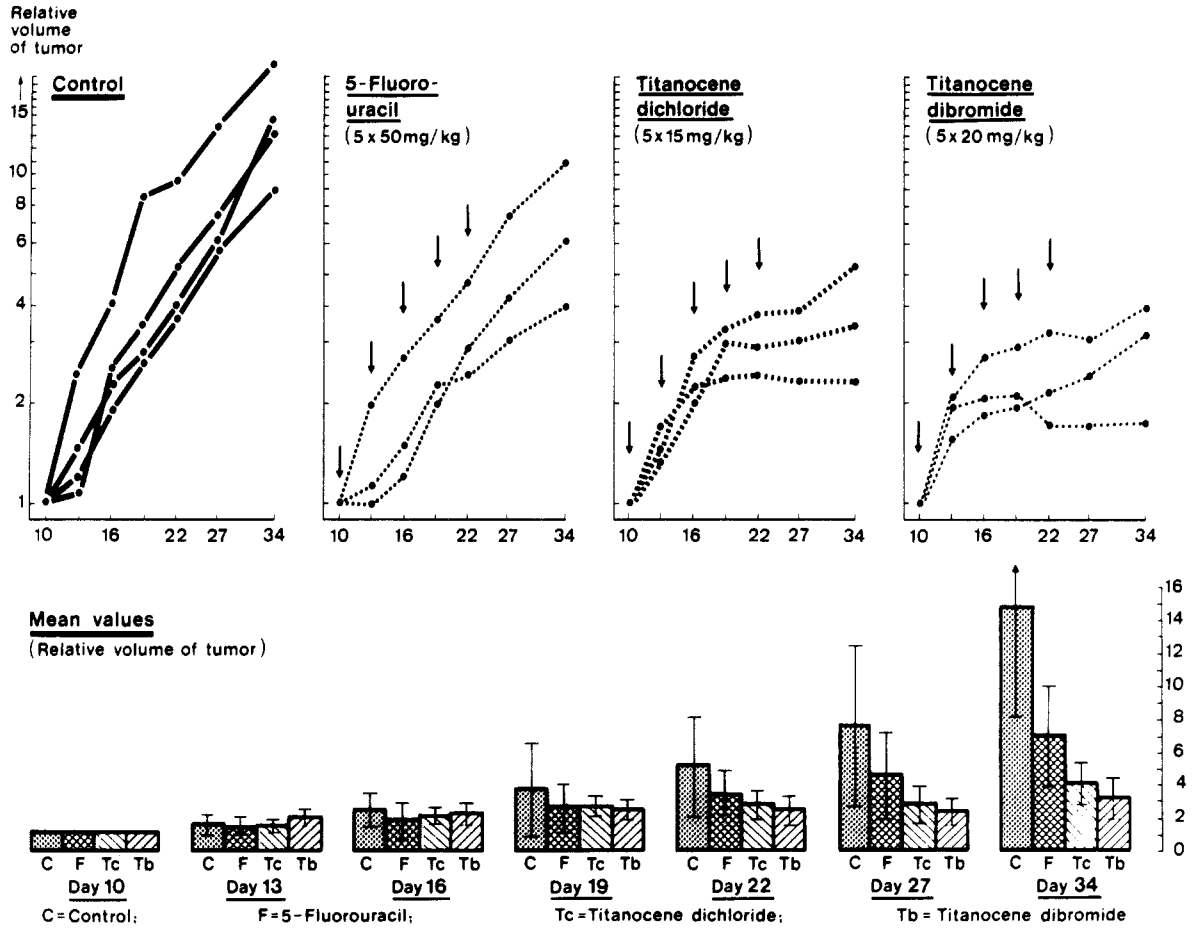


Figure 9. Growth development of a human colon adenocarcinoma heterotransplanted to athymic mice under treatment with 5-fluorouracil, **18** and **19**, administered at equivalent sublethal doses on days 10, 13, 16, 19, and 22 after tumor transplantation. Upper part: growth curves of individual tumors; abscissa, days after tumor implant on day 0; arrows indicate substance injections. Lower part: mean values of relative volume and standard deviations within control and treatment groups shown in the upper part.

parison purposes, 5-fluorouracil, a clinically approved cytostatic drug showing good activity especially against human colorectal carcinomas, was also tested at equitoxic doses. It becomes evident from growth curves as well as from mean values of relative tumor volume that no stronger tumor inhibition was caused by 5-fluorouracil than by titanocenes (Figure 9).

When titanocene compounds, e.g. **18** and **19**, were administered to animals bearing human breast carcinoma, which is characterized by rapid proliferation velocity in nude mice, again T/C ratios of about 30% were caused. The inhibitions of tumor growth were stable and remained constant 2 weeks beyond the end of treatment period (Figure 10).

In Figure 11, the influence of **18** and two derivatives containing hydrophilic ligands on the growth of a heterotransplanted human adenocarcinoma of the lung is shown. All three compounds clearly reduced tumor growth by 60–70% to T/C ratios of about 30%.[36] It is remarkable that these suppressions were again persistent and that the tumors even diminished in size after the end of treatment.

Optimum inhibition values effected by titanocene complexes **18–20** in human xenografts are summarized in Figure 12a. There was pronounced tumor-inhibiting activity against colon adenocarcinoma, breast carcinoma, and lung malignancies, whereas in analogy to the results against animal tumors superior activity was observed for **18**. Recent experiments performed with a greater number of individual colorectal carcinomas heterotransplanted to athymic mice confirmed the antitumor activity of **18** against this particular tumor type (Figure 12b). In the case of 9 out of 11 colorectal carcinomas, **18** effected growth inhibitions by 50–80%, resulting in T/C ratios of 50–20%.

Regarding the effects induced by various ferrocenium compounds **26–29**, there was good antiproliferative activity against heterotransplanted carcinomas of the colon sigmoideum as well as of the rectum.[37] Against these tumors, growth inhibition by 70–80% of control values was effected by most ferrocenium compounds (Figure 13). In the case of the rectal carcinoma, tumor inhibition caused by ferrocenium complexes was clearly stronger than after application of titanocene complexes. Moreover, the growth of lung adenocarcinoma was also significantly suppressed, whereby tumor inhibition was most pronounced after application of the tetrachloroferrate(III) and picrate derivatives **29** and **27** (Figure 13).

In summary, metallocene compounds, meaning titanocene complexes of type **12** as well as ferrocenium salts of type **14**, exhibited antitumor activity against various xenografted human tumors such as colorectal carcinomas and lung malignancies. These results are remarkable because it is known from comparative studies[38] that there is a high correlation between the response of human xenografts to chemotherapy and the clinical results obtained with the same drugs. Especially, the suppression of tumor growth in the case of several colorectal carcinomas is a strong hint to the susceptibility

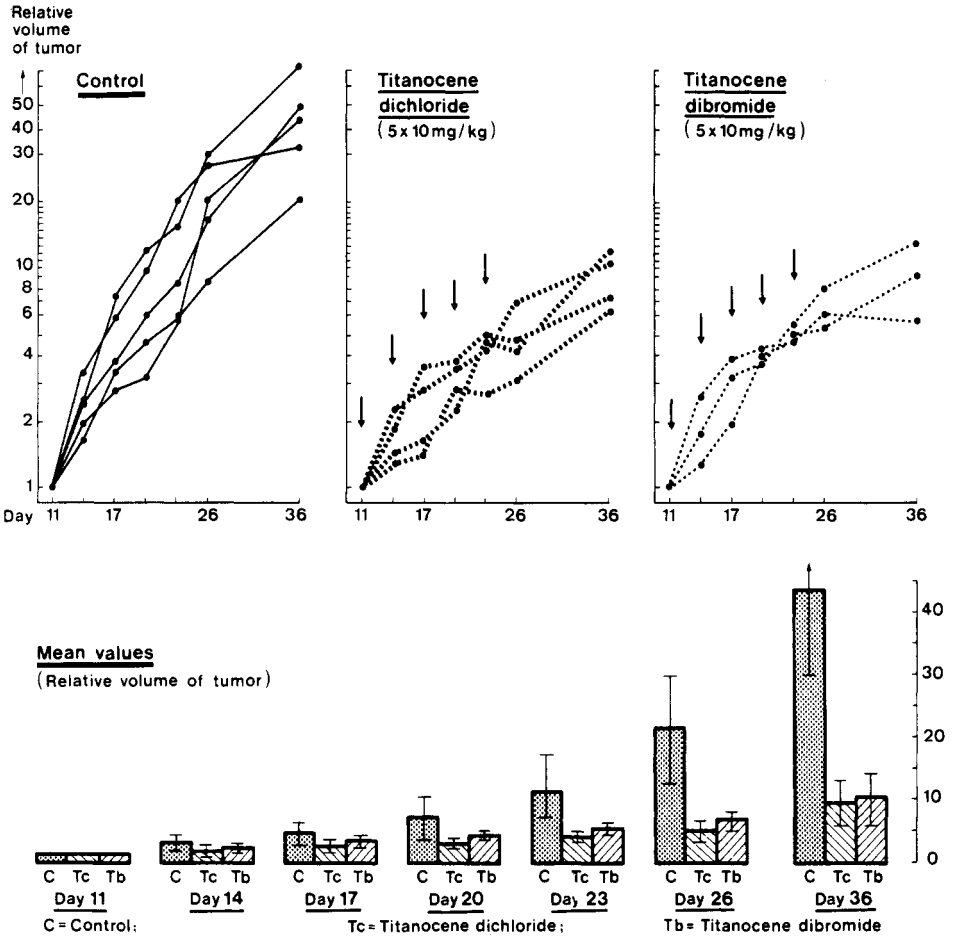


Figure 10. Growth development of a heterotransplanted human breast carcinoma under treatment with 18 and 19. For further details, cf. legend of Figure 9.

of this tumor type to 18. Regarding lung, breast, and cervix carcinomas, positive responses in a greater number of xenografted individual tumors are required to clearly underline the antitumor activity of 18 against these human tumor types.

V. Toxic Properties of Metallocene Complexes

Another important point for the evaluation and development of new cytostatic drugs is the problem of organ toxicity. Four questions must be considered: What is the dose-limiting organ toxicity? Does the pattern of toxicity resemble that of other cytostatics? Do the drugs severely damage bone marrow function? Is there embryotoxic and teratogenic influence in mammals?

These problems were investigated in mice after administration of single doses of $(C_5H_5)_2TiCl_2$ (18) at different dose levels (ED_{90}, 40 mg/kg; LD_{10}, 60 mg/kg). For comparison purposes, the inorganic cytostatic drug cisplatin (1) applied at the ED_{90} level (10 mg/kg) was used as control compound. The pattern of organ toxicity including myelotoxicity was analyzed by examination of blood and urine chemical parameters and by determination of peripheral hematologic counts.

It is known that 1 is burdened by severe nephrotoxicity,[39] which is manifested by structural lesions of proximal and distal tubular cells and by functional disturbances such as an increase of blood retention values or proteinuria and glucosuria. In contrast to these effects, no functional impairments of the kidneys were detectable after application of 18.[40] This is documented in Figure 14 for the values of blood urea nitrogen (BUN) and creatinine. Whereas after application of 1 significant elevations, especially of BUN values, manifested 1–16 days after treatment, no long-lasting increases were observable after treatment with 18 at the ED_{90} and LD_{10} levels. The short-lasting increase of creatinine in the serum immediately after application of 18 points to a transient impairment of renal blood perfusion, but not to a permanent renal lesion. After treatment with 18 or $(C_5H_5)_2VCl_2$ (22), no changes of urinary composition were found with respect to the content of proteins, glucose, nitrites, ketones, bilirubin, and urobilinogen. No appearance of corpuscular elements such as erythrocytes or leukocytes was detectable in the urine.[41]

Moreover, the histologic and ultrastructural analyses confirmed the apparent nonaffection of the kidneys by 18 and 22.[41,42] Neither epithelial cells of the proximal and distal tubules nor other cellular elements of the kidneys showed any pathologic lesions; their appearance was inconspicuous and identical with that of control kidneys even after application of an LD_{50} dose of 18 (Figures 15–18).

Whereas the kidneys were not injured even by toxic doses of 18, the serum levels of some liver enzymes, such as glutamate dehydrogenase (GLDH), glutamic-oxaloacetic transaminase (GOT), and glutamic-pyruvic transaminase (GPT), increased within 1 day after treatment with 18 to significantly elevated values (Figure 19), indicating an injury of the integrity of liver

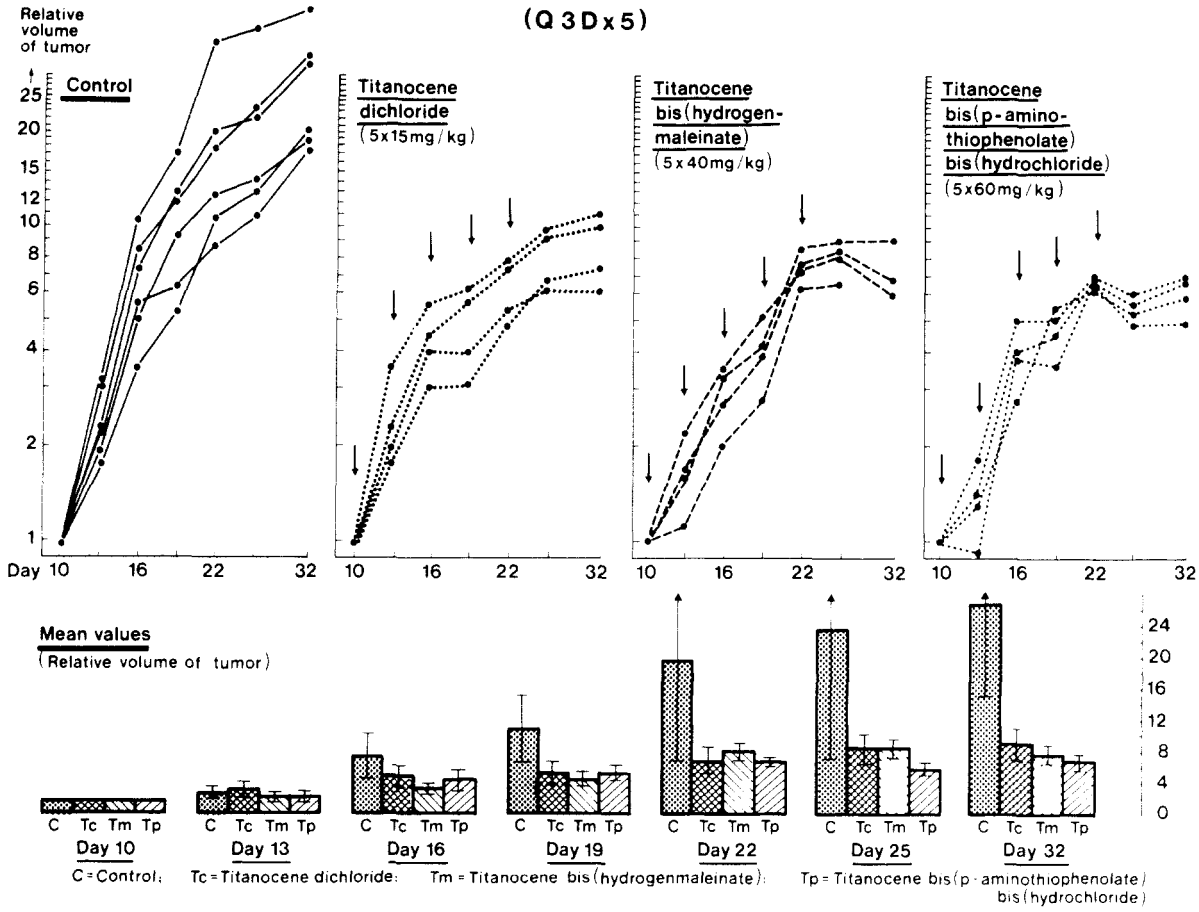


Figure 11. Growth development of a heterotransplanted human adenocarcinoma of the lung under treatment with various titanocene complexes, e.g. **18** and **20**. For further details, cf. legend of Figure 9.

cells.[40] It is worth mentioning that these findings were reversible and of transient character and that 8 days after substance injection the enzyme values of GLDH, GOT, and GPT had again normalized and were no longer elevated over control values.

These functional findings correlated to the morphologic appearance of liver cells after treatment with **18**. The occurrence of small lipid droplets in the cytoplasm of many cells of liver parenchyma within 1–2 days after substance application indicated ongoing fatty degeneration of liver cells. Single-cell necroses were detectable 8–12 days later.[34] It is known that similar phenomena usually develop under the influence of hepatotoxic agents. In analogy to the above-described functional impairments, the structural alterations of liver cells were also reversible and disappeared within 16–32 days after substance application.

The increased serum levels of liver enzymes are only explainable by disturbances of the structural integrity of liver cells. However, the excretion function of liver cells was obviously not injured; thus, the bilirubin and cholesterol serum levels remained unelevated during the whole experimental period.[40]

Organ function was influenced by application of titanocene compounds, in the endocrine glands such as the suprarenal cortex and the pancreas.[40] The serum content of some of the hormones produced in these organs, especially of cortisol and glucagon, markedly increased in mice by factors of 3–4 (Figure 20). Possibly, the initial decrease of serum glucose might be the stimulating factor for the forced output of both hormones.

Another interesting point is the problem of bone marrow depression, which mostly represents the dose-limiting toxicity of cytostatic drugs. In contrast to this general feature, **18** only slightly affected bone marrow function.[43] The counts of neither mature erythrocytes nor reticulocytes and polychromatophilic erythrocytes, which represent young red blood cells, decreased below control range at any time after application of **18**. On the other hand, in the case of **1**, the supply of young erythrocytes from bone marrow was markedly reduced near to zero level.

Comparable phenomena became obvious regarding the leukocyte count.[43] No depression in the case of **18** but a significant diminution of leukocytes in the peripheral blood below control values took place after application of **1** (Figure 21).

Finally, a decreased number of circulating platelets was detectable after application both of **1** and **18** (Figure 21). Thus, thrombocytes apparently represent the only cells affected by single doses of **18**.[43]

Toxic effect patterns induced by the antitumor agents **18** and **1** in mice are summarized in Figure 22. It becomes evident that the pattern of organ toxicity induced by **18** fundamentally differs from that induced by **1** as well as from those toxic features commonly found for organic cytostatic agents. There is no nephrotoxicity by **18**; dose-limiting toxicity seems to be an injury of liver cells. Bone marrow toxicity is remarkably low.

Generally, cytostatic drugs suppress and inhibit cellular proliferation processes by different mechanism. It is therefore comprehensible that most of them induce

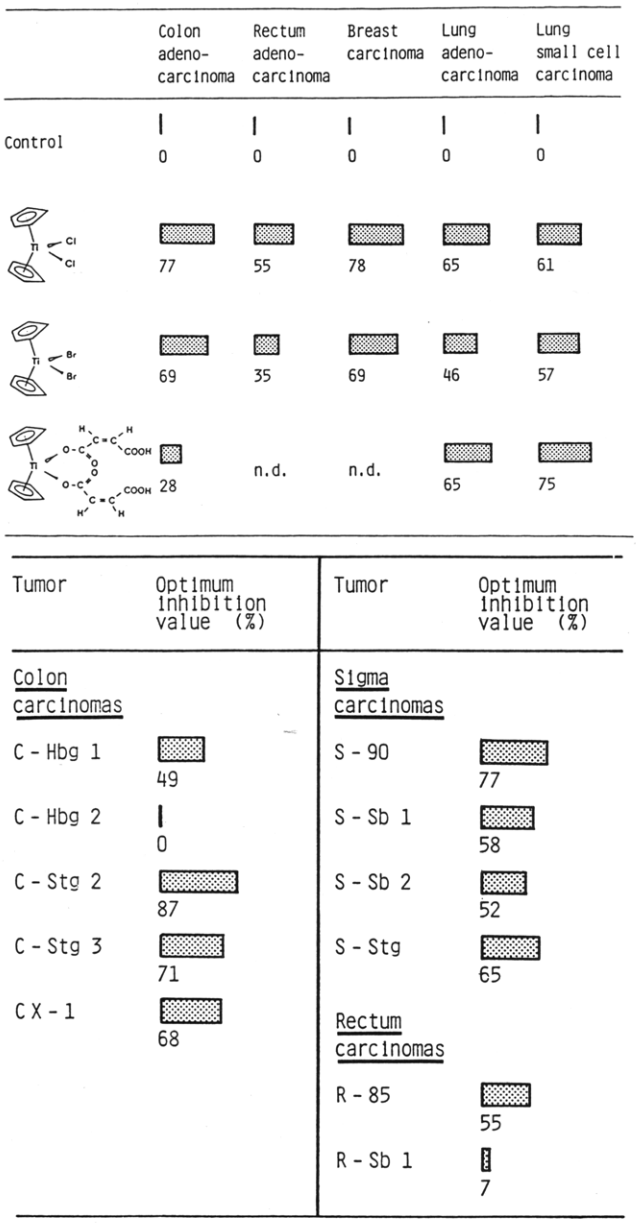


Figure 12. (Top, a) Growth inhibitions of human xenografts by titanocene complexes **18–20**. Optimum inhibition values are given in percent of control tumor size (100% − T/C) 3 or 4 days after last substance application. Substance application according to q3d × 5 or q2d × 5. Applied doses correspond to LD_{10} regimens. (Bottom, b) Growth inhibitions of 11 human colorectal carcinomas, heterotransplanted to athymic mice, after treatment with **18**.

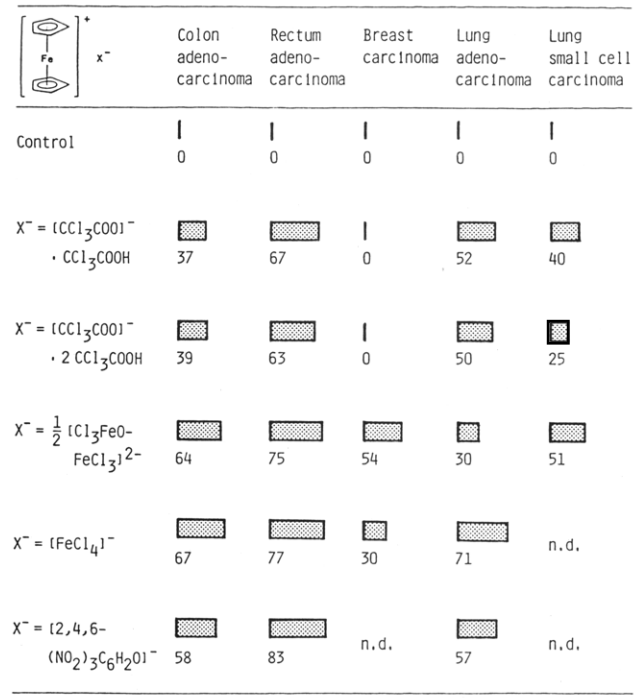


Figure 13. Optimum values of growth inhibition, effected in human xenografts by ferrocenium complexes **26–29**. For further details, cf. legend of Figure 12.

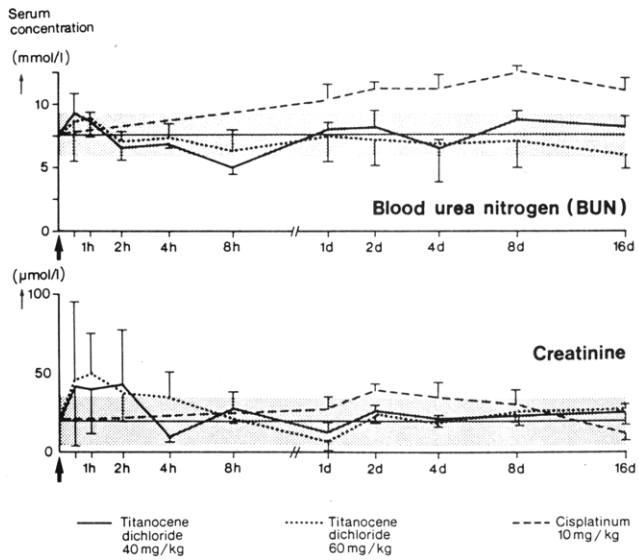


Figure 14. Serum content of blood urea nitrogen (BUN) and creatinine at various intervals (given on abscissa) after administration of **18** (40 or 60 mg/kg) or **1** (10 mg/kg) to mice on time 0. Given are mean values and standard deviations as $+s$ or $-s$. Straight continuous lines and shaded areas represent mean values and ranges of standard deviations of control populations.

severe malformations and growth retardations in developing embryos and fetuses when pregnant animals or women are treated with cytostatic agents.[44] Applying **18** to pregnant mice at various days between early organogenesis and late fetal period, however, led to surprising findings:[45] There was no appearance of multiple malformations in numerous organ systems, as is typical for other cytostatics such as alkylating agents, antimetabolites, or vinca rosea alkaloids. The only malformation found in 10–50% of the fetuses after application of **18** during organogenesis was the appearance of cleft palate (Figure 23). The occurrence of this symptom was a clearly dose- and time-dependent phenomenon.

In looking for the causes leading to the selective appearance of cleft palate, in principle, direct as well as indirect mechanisms may be responsible. The experiments performed in this connection pointed to indirect mechanisms to be responsible, as no titanium-containing species was found able to traverse the placental barrier to a mentionable extent.[34] Probably, the increase in the maternal serum content of glucocorticoids is the factor causing cleft palate in mice.[46] On the other hand, there do not exist any hints that glucocorticoids may also induce cleft palate during human gestation.[44b]

VI. Cellular and Molecular Mode of Action

Whereas, in the case of **1**, detailed conceptions about the probable intracellular mechanisms of action were

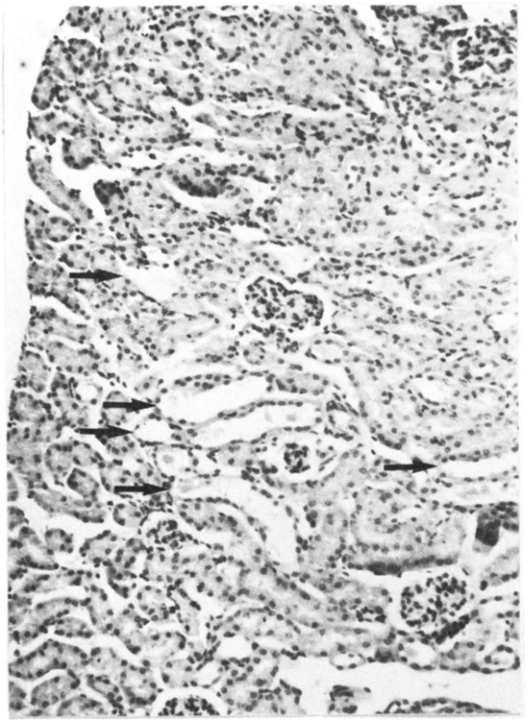


Figure 15. Histologic appearance of the renal cortex 8 days after application of **1** (10 mg/kg, ED_{90}). Many dilated tubules (→) and numerous necrotic tubular cells with condensed nuclei are seen after treatment with **1** (×150).

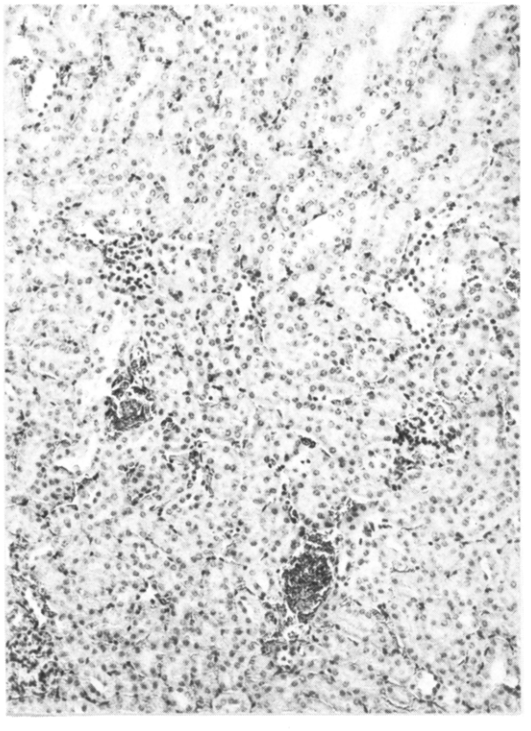


Figure 16. Histologic appearance of the renal cortex 8 days after application of **18** (100 mg/kg, LD_{50}). No pathologic changes after the application of LD_{50} dose of **18** (×150).

described and experimentally confirmed,[47] there exist only some indirect indications that the nucleic acid metabolism seems to be primarily disturbed by titanocene complexes. This became evident, for example, from incorporation studies with tritium-labeled precursors of the DNA, RNA, and protein syntheses.[48] These studies revealed (in vivo and in vitro) after application of **18** and **22** a persistent depression of DNA

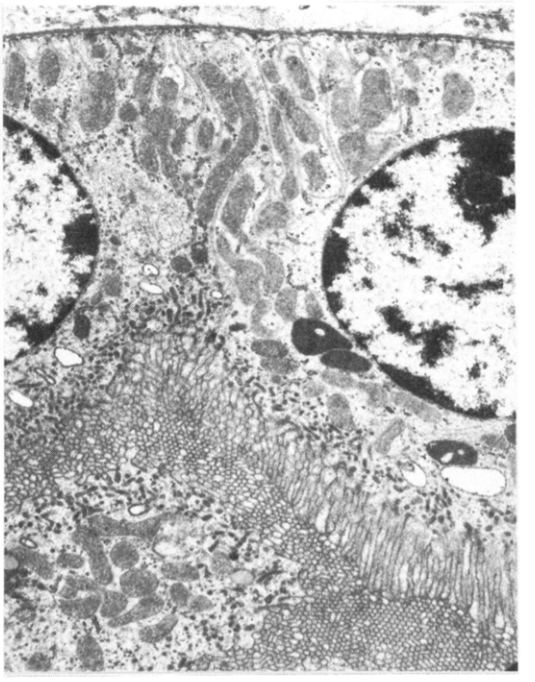


Figure 17. Part of a proximal convoluted tubule of the kidney 4 days after application of LD_{50} dose of **18** (100 mg/kg). The ultrastructural features of renal cells shown do not differ from those of untreated cells (×11 000).

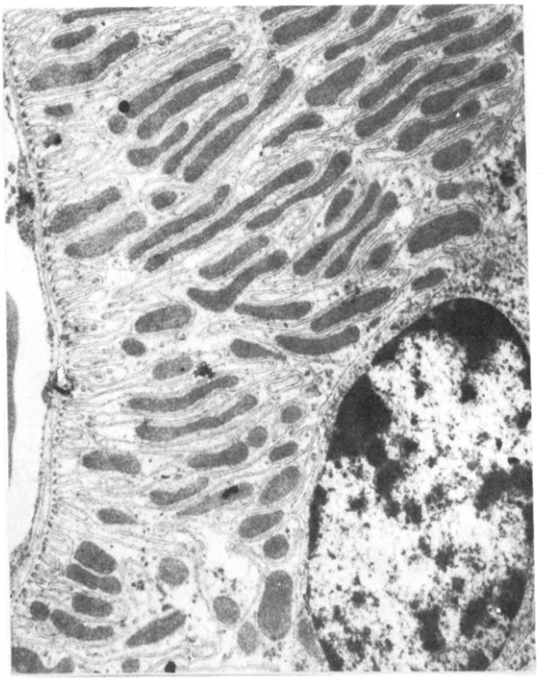


Figure 18. Part of a distal convoluted tubule of the kidney 4 days after application of LD_{50} dose of **22** (110 mg/kg). The ultrastructural features of renal cells shown do not differ from those of untreated cells (×11 000).

synthesis, whereas the RNA and the protein syntheses were inhibited only slightly and reversibly (Figure 24).

Analysis of the intracellular localization of titanium after treatment with **18** by use of electron energy loss spectroscopy[49] demonstrated that the central metal titanium mainly accumulated in those cellular regions rich in nucleic acids. Most titanium was enriched in the nuclear heterochromatin, smaller amounts were recognizable in the euchromatin, the nucleolus, and those cytoplasmic regions rich in ribosomes. A similar

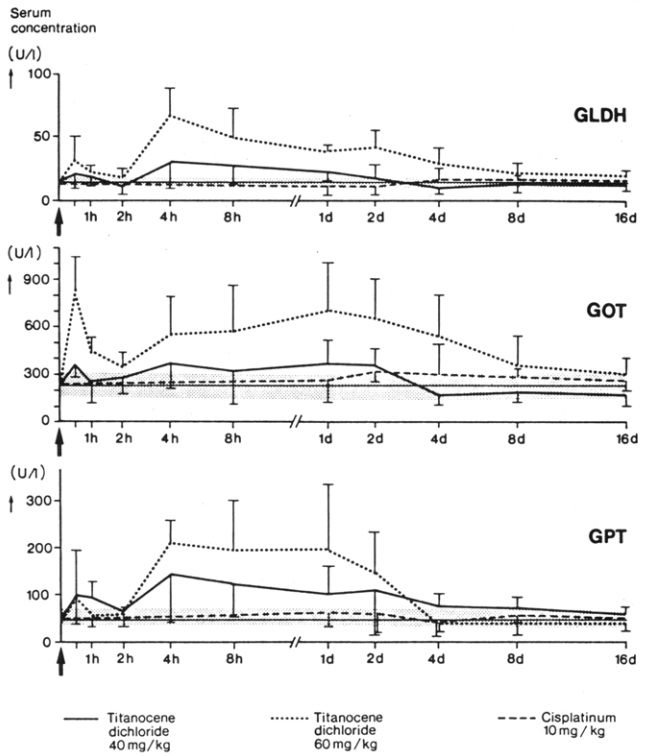


Figure 19. Serum content of some enzymes in the peripheral blood. For further explanations, cf. legend of Figure 14.

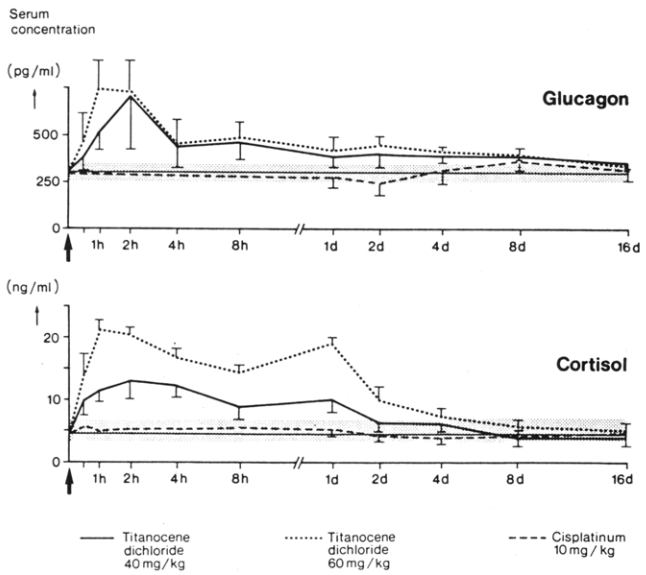


Figure 20. Serum content of some hormones involved in glucose regulation. For further explanations, cf. legend of Figure 14.

pattern of vanadium distribution was obtained after treatment with **22**.

The cytologic phenomena, observed in tumor cells after treatment with **18** and **22**, were in good agreement with the supposition that metallocene complexes primarily attack DNA molecules.[50] Three main developments were discernible within most tumor cells treated in vivo or in vitro (Figure 25): the formation of giant cells; the activation and morphological expression of endogenous viruses in the case of **18**; the development of cellular necrosis.

All these cytological phenomena are explainable by a molecular attack of metallocene dihalides upon intracellular DNA, and by this, they confirm the conception of primary attack upon nucleic acids.

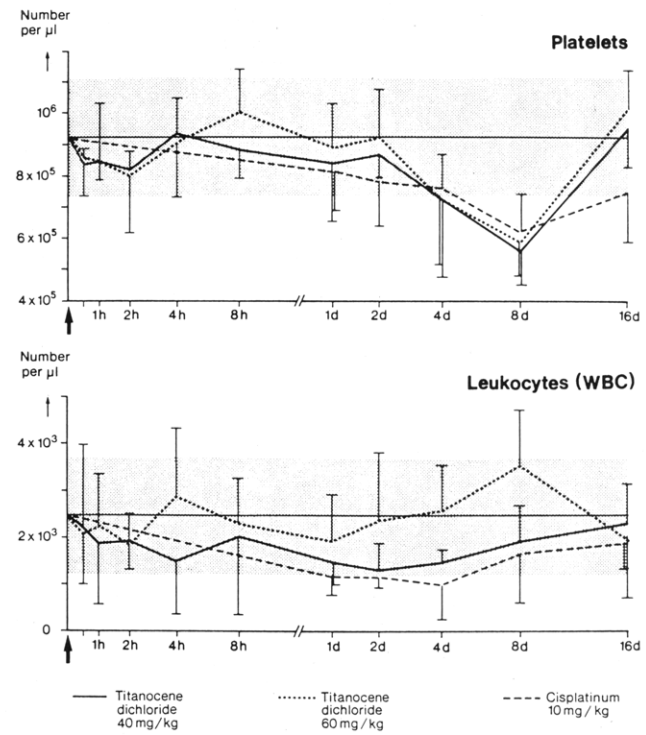


Figure 21. Counts of platelets (thrombocytes) and leukocytes in the peripheral blood. For further explanations, cf. legend of Figure 14.

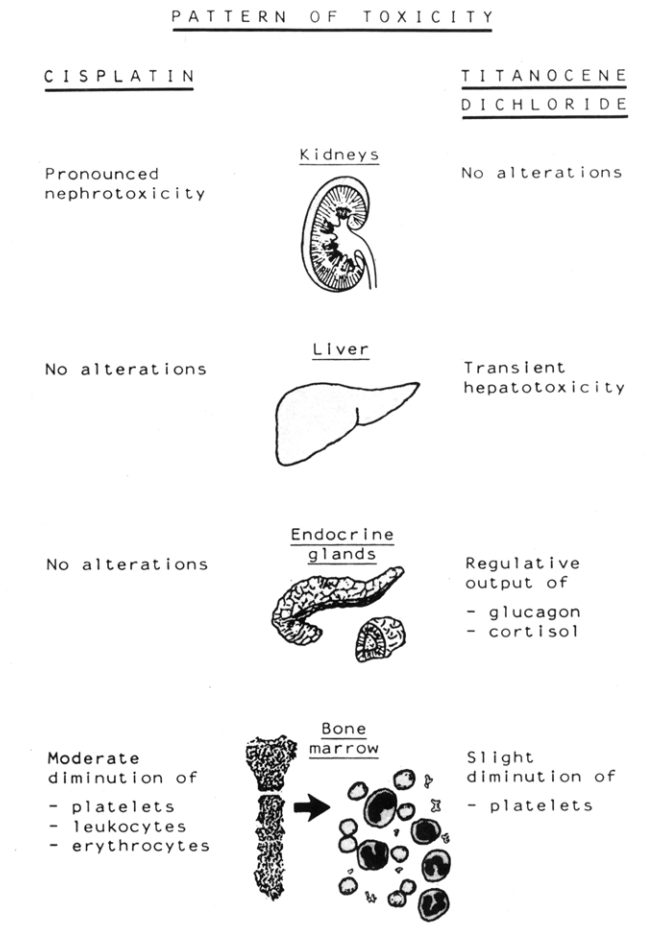


Figure 22. Comparison of main toxic effects induced in mice by **1** and **18**.

To gain insight into the molecular mode of action of metallocene dihalides, attempts have been made to synthesize model complexes of dicyclopentadienylmetal

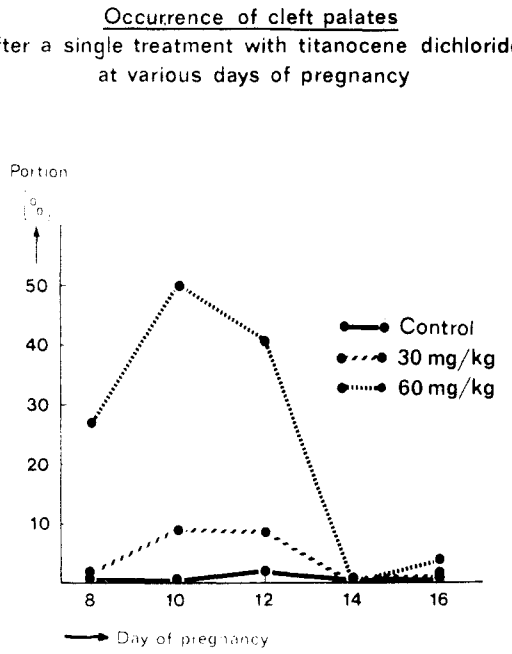


Figure 23. Portion of fetuses with cleft palate on day 18 of pregnancy after treatment of pregnant mice with 18.

moieties with nucleobases, nucleotides, and related molecules.

Cozak and co-workers[51] observed monofunctional bonding of a chlorodicyclopentadienyltitanium(IV) unit to the N-9 atom of purine (HPur) in $(C_5H_5)_2TiCl(Pur)$, as well as bifunctional chelation of dicyclopentadienyltitanium(III) units to the N-7 and O-6 atoms of theophylline (HThe) in $(C_5H_5)_2Ti(The)$ and to both the N-7, O-6 and the N-1, O-2 atoms of xanthine (H_3Xan) in the trinuclear $(C_5H_5)_2TiCl(HXan)[(C_5H_5)_2Ti]_2$, which additionally contains a monofunctional dative bond of N-9 to a chlorodicyclopentadienyltitanium(III) center. The value of these models is limited by the fact that the complexes have been obtained under nonphysiological conditions and some of them starting from the low-valent titanium species $[(C_5H_5)_2TiCl]_2$ or $(C_5H_5)_2Ti(CO)_2$. Marks and co-workers[52] investigated the aqueous coordination chemistry of **22** with nucleotides and phosphoesters and pointed to the coordination of $[(C_5H_5)_2V(OH_2)_2]^{2+}$ ions to phosphate groups of nucleotides, mediated by strong hydrogen bonds between aquo and phosphate oxygen atoms.

These results again show that the interactions of metallocene complexes with nucleic acids are possible and may actually be responsible for the antitumor activity of metallocenes. Nevertheless, the character of these interactions, which presumably depends on the linear metallocenium or pseudotetrahedral metallocene diligand type of complex, and, within these types, on the nature of the central metal atoms, seems to be more or less different from that described for 1.

The assessment of the actual importance of the mentioned model complexes with regard to the biological mode of action is rendered more difficult by the fact that, in aqueous solution, most of the metallocene complexes are not stable but undergo dissociation, aquation, and hydrolysis reactions. In a kinetic study published by Marks and co-workers,[53] the order of decreasing hydrolytic stability of the $M-C_5H_5$ bond of metallocene dichlorides in unbuffered aqueous KNO_3 solution was determined to be $(C_5H_5)_2VCl_2$ (**22**) > $(C_5H_5)_2TiCl_2$ (**18**) ≫ $(C_5H_5)_2ZrCl_2$. Dissociation of the first chloride ion was found to be too rapid to be measured. Approximate half-lives for the loss of the second chloride amounted to 50 min for **18**, 30 min for $(C_5H_5)_2ZrCl_2$, and 24 min for **22**, the equilibrium constants being $K_2 = 4.2 \times 10^{-2}$ (**18**) and 2.7×10^{-3} (**22**).

Probably, this apparent higher hydrolytic stability of the $(C_5H_5)_2M^{2+}$ framework in **22** may be responsible for the much more pronounced growth-inhibiting potencies of **22** in vitro in comparison to **18**.[54] Because, on the other hand, **22** and **18** effect both equivalent antitumor activity in vivo, the results point to possible stabilizing interactions between metallocenes and blood constituents, e.g. proteins and lipid molecules, which have not been considered in the study of Marks and co-workers.[53] Because of the higher hydrolytic stability of the $(C_5H_5)_2V^{2+}$ framework at physiological pH, these au-

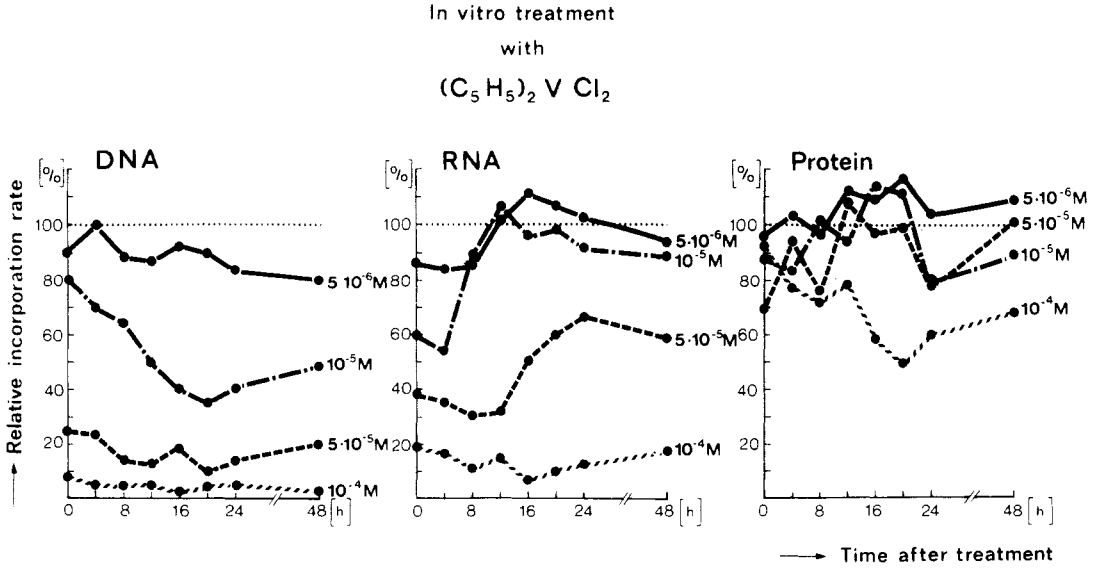


Figure 24. Incorporation rates, related as percentage to controls (···), of [*methyl*-^{3}H]thymidine (left), [5-^{3}H]uridine (middle), and L-[4,5-^{3}H]leucine (right) as a measure of DNA, RNA, and protein synthesis activities, respectively, after a 90-min treatment of Ehrlich ascites tumor cells with **22** in vitro.

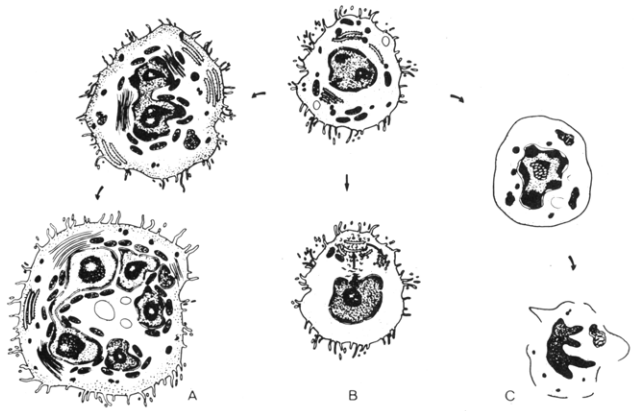


Figure 25. Schematic representation of main morphological phenomena within Ehrlich ascites tumor cells growing in vitro during and after exposure to **18** or **22**: a, cellular enlargement and giant cell formation; b, activation and morphological expression of viruses after exposure; c, cellular degeneration and development of cellular necrosis.

thors concluded[52,53] that **22** should be the compound of choice for further mechanistic and antitumor studies. In this connection, it is worth mentioning that it was possible during the past months to develop an appropriate galenic preparation preventing hydrolytic reactions of **18**. Interestingly, the antitumor potential of **18** was unchanged and preserved under these conditions.[34]

VII. Conclusions

The following conclusions can be drawn from the experimental results known about non-platinum-group metal antitumor agents:

(1) Non-platinum-group metal antitumor agents are represented by different groups of inorganic and organometallic compounds containing either main-group elements such as Ga, Ge, and Sn or early, medium, and late transition metals such as Ti, V, Fe, Cu, or Au.

(2) Most of these compounds are characterized by spectra of activity against experimental animal and human tumors not identical with those known from platinum complexes. It may be expected that these differences considering the preclinical spectrum of activity are indicative of differences in the clinical spectrum of activity and that there does not exist cross-resistance between platinum and non-platinum-group metal cytostatics.

(3) The toxic properties of most non-platinum-group metal cytostatic agents, especially of those containing less heavy metals such as Ti or Fe, fundamentally differ from those of platinum compounds. Theoretically, this constellation opens the possibility of combination therapy without the danger that toxic properties will potentiate.

(4) From these biological features, it can be concluded that non-platinum-group metal antitumor agents do not represent analogues of antitumor platinum complexes but that they must be considered as independent groups of antiproliferative agents.

VIII. Perspectives

The recent detections of various, quite differing non-platinum-group metal antitumor agents justify the assessment that we are presently only at the beginning of a new development and that, probably, numerous novel results will be uncovered during the next years.

It may be expected that other cytostatically active complexes of Ti, V, Fe, Cu, Au, Ga, Ge, and Sn will be detected in the near future and that further antitumor compounds containing non-platinum-group metals differing from these elements will be found.

This means that inorganic and organometallic complexes obviously represent an arsenal of substances that are actually only sporadically explored with respect to cytostatic properties. The antiproliferative potencies that have been shown for some of them during the past few years qualify them as promising compounds for further antitumor studies.

Moreover, it seems probable that some of these compounds will also exhibit pharmacological properties different from antitumor activity. In pilot experiments, for example, antiviral, antiinflammatory, and antiarthritic activities have been revealed for titanocene dichloride. The search for unusual pharmaceuticals therefore represents a fascinating and important topic in the field of bioinorganic and bioorganometallic chemistry.

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X. References

(1) Rosenberg, B.; Van Camp, L.; Trosko, J. E.; Mansour, V. H. *Nature (London)* **1969**, *222*, 385.
(2) (a) Cleare, M. J.; Hydes, P. C.; Hepburn, D. R.; Malerbi, B. W. In *Cisplatin: Current Status and New Developments*; Academic: New York, 1980. (b) Wolpert-DeFilippes, M. K. In Reference 2a. (c) Harrap, K. R.; Jones, M.; Wilkinson, C. R.; Clink, H. M.; Sparrow, S.; Mitchley, B. C. V.; Clarke, S. In Reference 2a. (d) Roberts, J. J. In *Metal Ions in Genetic Information Transfer*; Eichhorn, G. L., Marzilli, L. G., Eds.; Elsevier: New York, 1981.
(3) (a) Kelson, D.; Scher, H.; Burchenal, J. *Dev. Oncol.* **1984**, *17*, 310. (b) Calvert, A. H.; Harrap, K. R.; Wiltshaw, E.; Smith, I. E. *Dev. Oncol.* **1984**, *17*, 240.
(4) Weiss, G. R.; Kisner, D. L.; Kuhn, J. G.; Melink, T. J.; Myers, J. W.; Von Hoff, D. D. In *Cancer Chemotherapy Annual* 6; Pinedo, H. M., Chabner, B. A., Eds., Elsevier: New York, 1984.
(5) Bear, J. L.; Gray, H. B.; Rainen, L.; Chang, I. M.; Howard, R.; Serio, G.; Kimball, A. P. *Cancer Chemother. Rep. Part 1* **1975**, *59*, 611.
(6) (a) Giraldi, S.; Sava, G.; Bertoli, G.; Mestroni, G.; Zassinovich, G. *Cancer Res.* **1977**, *37*, 2662. (b) Clarke, M. J. In *Inorganic Chemistry in Biology and Medicine*; Martell, E. A., Ed.; American Chemical Society: Washington, DC, 1980.
(7) (a) Gill, D. S. *Dev. Oncol.* **1984**, *17*, 267. (b) Graham, R. D.; Williams, D. R. *J. Inorg. Nucl. Chem.* **1979**, *41*, 1245.
(8) Sava, G.; Giraldi, T.; Mestroni, G.; Zassinovich, G. *Chem.Biol. Interact.* **1983**, *45*, 1.
(9) Adamson, R. H.; Canellos, G. P.; Sieber, S. M. *Cancer Chemother. Rep. Part 1* **1975**, *59*, 599.
(10) Bedikian, A. Y.; Valdivieso, M.; Bodey, G. P.; Burgess, M. A.; Benjamin, R. S.; Hall, S.; Freireich, E. J. *Cancer Treat. Rep.* **1978**, *62*, 1449.
(11) (a) Mulinos, M. G.; Amin, P. *Fed. Proc. Fed. Am. Soc. Exp. Biol.* **1980**, *39*, 747. (b) Schein, P. S.; Slavik, M.; Smythe, T.; Hoth, D.; Smith, F.; MacDonald, J. S.; Wooley, P. V. *Cancer Treat. Rep.* **1980**, *64*, 1051.
(12) (a) Kumano, N.; Nakai, Y.; Ishikawa, T.; Koinumaru, S.; Suzuki, S.; Konno, K. *Sci. Rep. Res. Inst. Tohoku Univ., Ser. A* **1978**, *25*, 89. (b) Kumano, N.; Nakai, Y.; Ishikawa, S.; Koinumaru, S.; Suzuki, S.; Kikumoto, T.; Konno, K. Proceedings of the 19th Interscience Conference on Antimicrobial Agents and Chemotherapy, 1979, No. 1004.
(13) (a) Aso, H.; Suzuki, F.; Yamaguchi, T.; Hayashi, Y.; Ebina, T.; Ishida, N. *Jpn. J. Cancer Chemother.* **1982**, *9*, 1976. (b) Suzuki, F.; Brutkiewicz, R. R.; Pollard, R. B. Anticancer Res. **1985**, *5*, 479. (c) Badger, A. M.; Mirabelli, C. K.; DiMartino, M. *Immunopharmacology* **1985**, *10*, 201.
(14) Crowe, A. J.; Smith, P. J.; Atassi, G. *Chem.-Biol. Interact.* **1980**, *32*, 171.

(15) (a) Barbieri, R.; Pellerito, L.; Ruisi, G.; LoGiudice, M. T.; Huber, F.; Atassi, G. *Inorg. Chim. Acta* **1982**, *66*, L 39. (b) Huber, F. Proceedings of the 4th International Conference on Organometallic Coordination Chemistry of Ge, Sn, and Pb, McGill University, Montreal, Canada, 1983. (c) Haiduc, I.; Silvestru, C.; Gielen, M. *Bull. Soc. Chim. Belg.* **1983**, *92*, 187. (d) Saxena, A.; Tandon, J. P. *Cancer Lett. (Shannon, Irel.)* **1983**, *19*, 73. (e) Huber, F.; Roge, G.; Carl, L.; Atassi, G.; Spreafico, F.; Filippeschi, S.; Barbieri, R.; Silvestri, A.; Rivatola, E.; Ruisi, G.; DiBianca, F.; Alonzo, G. *J. Chem. Soc., Dalton Trans.* **1985**, 523.
(16) Crowe, A. J.; Smith, P. J.; Atassi, G. *Inorg. Chim. Acta* **1984**, *93*, 179.
(17) (a) Köpf, H.; Köpf-Maier, P. *Angew. Chem.* **1979**, *91*, 509; *Angew. Chem., Int. Ed. Engl.* **1979**, *18*, 477. (b) Köpf-Maier, P.; Köpf, H. *Z. Naturforsch., B: Anorg. Chem., Org. Chem.* **1979**, *34B*, 805. (c) Köpf-Maier, P.; Leitner, M.; Voigtländer, R.; Köpf, H. *Z. Naturforsch., C: Biosci.* **1979**, *34C*, 1174. (d) Köpf-Maier, P.; Leitner, M.; Köpf, H. *J. Inorg. Nucl. Chem.* **1980**, *42*, 1789. () Köpf, H.; Köpf-Maier, P. *ACS Symp. Ser.* **1983**, *No. 209*, 315.
(18) (a) Köpf-Maier, P.; Preiss, F.; Marx, T.; Klapötke, T.; Köpf, H. *Anticancer Res.* **1986**, *6*, 33. (b) Köpf-Maier, P.; Köpf, H. *Arzneim.-Forsch.* **1987**, *37*, 532. (c) Köpf-Maier, P.; Moormann, A.; Köpf, H. *Eur. J. Cancer Clin. Oncol.* **1985**, *21*, 853. (d) Köpf-Maier, P., unpublished results.
(19) (a) Keller, H. J.; Keppler, B.; Schmähl, D. *Arzneim.-Forsch.* **1982**, *32*, 806. (b) Keller, H. J.; Keppler, B.; Schmähl, D. *J. Cancer Res. Clin. Oncol.* **1983**, *105*, 109. (c) Keppler, B. K.; Diez, A.; Seifried, V. *Arzneim.-Forsch.* **1985**, *35*, 1832.
(20) Keppler, B. K.; Michels, K. *Arzneim.-Forsch.* **1985**, *35*, 1837.
(21) (a) Köpf-Maier, P.; Köpf, H.; Neuse, E. W. *Angew. Chem.* **1984**, *96*, 446; *Angew. Chem., Int. Ed. Engl.* **1984**, *23*, 456. (b) Köpf-Maier, P.; Köpf, H.; Neuse, E. W. *J. Cancer Res. Clin. Oncol.* **1984**, *108*, 336.
(22) (a) Antholine, W. E.; Kalyanaraman, B.; Petering, D. H. *EHP, Environ. Health Perspect.* **1985**, *64*, 19. (b) Bushnell, G. W.; Tsang, A. Y. M. *Can. J. Chem.* **1979**, *57*, 603.
(23) Elo, H. O.; Lumme, P. O. *Cancer Treat. Rep.* **1985**, *69*, 1021.
(24) Sadler, P. J.; Nasr, M.; Narayanan, V. L. *Dev. Oncol.* **1984**, *17*, 290.
(25) (a) Berghof, F. E.; Berghof, K.; Walz, D. T. *J. Rheumatol.* **1978**, *5*, 68. (b) Capell, H. A., Cole, D. S., Manghani, K. K., Morris, R. W., Eds. *Auranofin*, Proceedings of the Smith Kline and French International Symposium; Excerpta Medica: Amsterdam, 1983.
(26) (a) Simon, T. M.; Kunishima, D. H.; Vibert, G. J.; Lorber, A. *Cancer Res.* **1981**, *41*, 94. (b) Mirabelli, C. K.; Johnson, R. K.; Sung, C. M.; Faucette, L.; Muirhead, K.; Crooke, S. T. *Cancer Res.* **1985**, *45*, 32.
(27) Shaw, C. F., III.; Beery, A.; Stocco, G. C. *Inorg. Chim. Acta* **1986**, *123*, 213.
(28) (a) Berners-Price, S. J.; Mirabelli, C. K.; Johnson, R. K.; Mattern, M. R.; McCabe, F. L.; Faucette, L. F.; Sung, C.-M.; Mong, S.-M.; Sadler, P. J.; Crooke, S. T. *Cancer Res.* **1986**, *46*, 5486. (b) Berners-Price, S. J.; Sadler, P. J. *Inorg. Chem.* **1986**, *25*, 3822.
(29) Köpf-Maier, P.; Hesse, B.; Köpf, H. *J. Cancer Res. Clin. Oncol.* **1980**, *96*, 43.
(30) Köpf-Maier, P.; Hesse, B.; Voigtländer, R.; Köpf, H. *J. Cancer Res. Clin. Oncol.* **1980**, *97*, 31.
(31) Köpf-Maier, P.; Grabowski, S.; Liegener, J.; Köpf, H. *Inorg. Chim. Acta* **1985**, *108*, 99.
(32) Toney, J. H.; Rao, L. N.; Murthy, M. S.; Marks, T. J. *Breast Cancer Res. Treat.* **1985**, *6*, 185.
(33) Köpf-Maier, P.; Wagner, W.; Hesse, B.; Köpf, H. *Eur. J. Cancer* **1981**, *17*, 665.
(34) Köpf-Maier, P. to be submitted for publication.
(35) (a) Köpf-Maier, P.; Kahl, W.; Klouras, N.; Hermann, G.; Köpf, H. *Eur. J. Med. Chem. Chim. Ther.* **1981**, *16*, 275. (b) Köpf-Maier, P.; Grabowski, S.; Köpf, H. *Eur. J. Med. Chem. Chim. Ther.* **1984**, *19*, 347.
(36) Köpf-Maier, P. to be submitted for publication.
(37) Köpf-Maier, P. Proceedings—Seminar on the Human Tumour Xenografts Anticancer Drug Development, Milano, Italy, 1986.
(38) (a) Fiebig, H. H.; Schuchhardt, C.; Heuss, H.; Fiedler, L.; Löhr, G. W. *Behring Inst. Mitt.* **1984**, *74*, 343. (b) Bellet, R. E.; Danna, V.; Mastrangelo, M. J.; Berd, D. *JNCI, J. Natl. Cancer Inst.* **1979**, *63*, 1185. (c) Fujita, M.; Hayata, S.; Taguchi, T. *J. Surg. Oncol.* **1980**, *15*, 211. (d) Steel, G. G.; Courtenay, V. D.; Peckham, M. J. *Br. J. Cancer* **1983**, *47*, 1.
(39) (a) Kociba, R. J.; Sleight, S. D. *Cancer Chemother. Rep. Part 1* **1971**, *55*, 1. (b) Schaeppi, U.; Heymann, J. A.; Fleischman, R. W.; Rosenkrantz, H.; Jlievski, V.; Phelan, R.; Cooney, D. A.; Davis, R. D. *Toxicol. Appl. Pharmacol.* **1973**, *25*, 230. (c) Bitran, J. D.; Desser, R. K.; Billings, A. A.; Kozloff, M. F.; Shapiro, C. M. *Cancer* **1982**, *49*, 1784.
(40) (a) Köpf-Maier, P.; Gerlach, S. *J. Cancer Res. Clin. Oncol.* **1986**, *111*, 243. (b) Köpf-Maier, P.; Köpf, H. *Anticancer Res.* **1986**, *6*, 227.
(41) Köpf-Maier, P.; Funke-Kaiser, P. *Toxicology* **1986**, *38*, 81.
(42) Köpf-Maier, P.; Funke-Kaiser, P.; Greiff, D. *Verh. Anat. Ges.* **1984**, *78*, 603.
(43) Köpf-Maier, P.; Gerlach, S. *Anticancer Res.* **1986**, *6*, 235.
(44) (a) Scott, W. J. In *Handbook of Teratology*; Wilson, J. G., Frazer, F. C., Eds.; Plenum: New York, 1978; Vol. 2. (b) Neubert, D.; Barrach, H.-J.; Merker, H.-J. In *Drug-Induced Pathology*; Grundmann, E., Ed.; Springer: New York, 1980.
(45) Köpf-Maier, P.; Erkenswick, P. *Toxicology* **1984**, *33*, 171.
(46) Köpf-Maier, P. *Toxicology* **1985**, *37*, 111.
(47) (a) Barton, J. K.; Lippard, S. J. *Ann. N.Y. Acad. Sci.* **1978**, *313*, 686. (b) Poirier, M. C.; Lippard, S. J.; Zwelling, L. A.; Ushay, H. M.; Kerrigan, D.; Thill, C. C.; Santella, R. M.; Grunberger, D.; Yuspa, S. H. *Proc. Natl. Acad. Sci. U.S.A.* **1982**, *79*, 6443. (c) den Hartog, J. H. J.; Altona, C.; Chottard, J.-C.; Girault, J.-P.; Lallemand, J.-Y.; de Leeuw, F. A. A. M.; Marcelis, A. T. M.; Reedijk, J. *Nucleic Acids Res.* **1982**, *10*, 4715. (d) Plooy, A. C. M.; Fichtinger-Schepman, A. M. J.; Schutte, H. H.; van Dijk, M.; Lohman, P. H. M. *Carcinogenesis* **1985**, *6*, 516. (e) Fraval, H. N. A.; Roberts, J. J. *Cancer Res.* **1979**, *39*, 1793.
(48) (a) Köpf-Maier, P.; Köpf, H. *Naturwissenschaften* **1980**, *67*, 415. (b) Köpf-Maier, P.; Wagner, W.; Köpf, H. *Naturwissenschaften* **1981**, *68*, 272.
(49) (a) Köpf-Maier, P.; Krahl, D. *Naturwissenschaften* **1981**, *68*, 273. (b) Köpf-Maier, P.; Krahl, D. *Chem.-Biol. Interact.* **1983**, *44*, 317.
(50) (a) Köpf-Maier, P. *J. Cancer Res. Clin. Oncol.* **1982**, *103*, 145. (b) Köpf-Maier, P.; Köpf, H. *Dev. Oncol.* **1984**, *17*, 279.
(51) (a) Beauchamp, A. L.; Cozak, D.; Mardhy, A. *Inorg. Chim. Acta* **1984**, *92*, 191. (b) Beauchamp, A. L.; Bélanger-Gariépy, F.; Mardhy, A.; Cozak, D. *Inorg. Chim. Acta* **1986**, *124*, 23. (c) Cozak, D.; Mardhy, A.; Olivier, M. J.; Beauchamp, A. L. *Inorg. Chem.* **1986**, *25*, 2600.
(52) Toney, J. H.; Brock, C. P.; Marks, T. J. *J. Am. Chem. Soc.* **1986**, *108*, 7263.
(53) Toney, J. H.; Marks, T. J. *J. Am. Chem. Soc.* **1985**, *107*, 947.
(54) Köpf-Maier, P.; Wagner, W.; Köpf, H. *Cancer Chemother. Pharmacol.* **1981**, *5*, 237.
(55) In this paper the periodic group notation is in accord with recent actions by IUPAC and ACS nomenclature committees. A and B notation is eliminated because of wide confusion. Groups IA and IIA become groups 1 and 2. The d-transition elements comprise groups 3 through 12, and the p-block elements comprise groups 13 through 18. (Note that the former Roman number designation is preserved in the last digit of the new numbering: e.g., III → 3 and 13.)