Pharmacokinetics and adverse reactions of a new liposomal cisplatin (Lipoplatin): Phase I study

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Abstract. Lipoplatin, a new liposomal cisplatin formulation, is formed from cisplatin and liposomes composed of dipalmitoyl phosphatidyl glycerol (DPPG), soy phosphatidyl choline (SPC-3), cholesterol and methoxy-polyethylene glycol-distearoyl phosphatidylethanolamine (mPEG2000-DSPE). Following intravenous infusion, the nanoparticles (110 nm) are distributed into tissues and concentrate preferentially at tumor sites supposedly via extravasation through the leaky tumor vasculature. This study was designed to investigate the pharmacokinetics and the toxicity of this new liposomal cisplatin in patients with pretreated advanced malignant tumors. The drug was infused for 8 h every fourteen days at escalating doses. Twenty-seven patients were included and 3-5 patients were selected for each dosage level; levels started at 25 mg/m^2 and were increased by 25 to 125 mg/m^2. Three patients were also treated at higher dose levels, one each at 200, 250 and 300 mg/m^2. Blood was taken at certain time intervals in order to estimate total platinum plasma levels. At level 5 (125 mg/m^2), grades 1 and 2 GI tract and hematological toxicities were detected. No nephrotoxicity was observed. Seven additional patients were added at the 4th level (100 mg/m^2 for further pharmacokinetic evaluation. Measurement of platinum levels in the plasma of patients as a function of time showed that a maximum platinum level is attained at 6-8h. The half-life of Lipoplatin was 60-117 h depending on the dose. Urine excretion reached about 40% of the infused dose in 3 days. The data demonstrate that Lipoplatin up to a dose of 125 mg/m^2 every 14 days has no nephrotoxicity and it lacks the serious side effects of cisplatin.

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Key words: Lipoplatin, toxicity, chemotherapy

Introduction

Cisplatin, and later its analogues such as carboplatin, have been milestone achievements in clinical oncology (1). Cisplatin is one of the most widely used and most effective cytotoxic agents in the treatment of epithelial malignancies such as lung, head-and-neck, ovarian, bladder and testicular cancers (2-12). However, continued clinical use has been impeded by severe adverse reactions including renal toxicity, gastrointestinal toxicity, peripheral neuropathy, asthenia, and ototoxicity (3,4). The significant risk of nephrotoxicity caused by cisplatin frequently hinders the use of higher doses to maximize its antineoplastic effects (13,14). Cisplatin when combined with other cytotoxic agents, has shown an improved response rate and survival in a moderate to high number of patients suffering from the above malignancies; it is not particularly myelotoxic but nephrotoxicity is often unacceptable. Cisplatin analogues have been marketed (carboplatin, oxaliplatin) but none as yet has achieved a similar broad-spectrum effectiveness.

Lipoplatin, a liposomal formulation of cisplatin, was developed in order to reduce the systemic toxicity of cisplatin while simultaneously improving the targeting of the drug to the primary tumor and to metastases by enhancing the circulation time in body fluids and tissues. Preclinical studies have shown Lipoplatin's lower nephrotoxicity in rats, as compared to cisplatin, the plasma pharmacokinetics and therapeutic efficacy in mouse xenografts with breast and prostate human tumors. The data are consistent with an apoptotic death of tumor cells after treatment of xenografts with Lipoplatin (15). Lipoplatin showed reduced renal toxicity in mice and rats; whereas animals injected with cisplatin developed renal insufficiency with clear evidence of tubular damage, those injected with the same dose of Lipoplatin were almost completely free of kidney injury (16). The objectives of the present Phase I study were to investigate the pharmacokinetics and toxicity of liposomal cisplatin.

Materials and methods

Preparation and characteristics of Lipoplatin. Cisplatin was purchased from Heraeus /Flavine (mw 300). The lipid shell of Lipoplatin is composed of 1,2-DiPalmitoyl-sn-Glycero-3-[Phospho-rac-(1-glycerol)] (sodium salt) also known as dipalmitoyl phosphatidyl glycerol (DPPG, mw 745, Lipoid GmbH), soy phosphatidyl choline (SPC-3, mw 790 Lipoid GmbH), cholesterol (CHOL, mw 386.66, Avanti Polar Lipids) and methoxy-polyethylene glycol-distearoyl phosphatidyl-ethanolamine lipid conjugate (mPEG2000-DSPE, mw 2807, Genzyme). Lipoplatin is composed of: 8.9% cisplatin and 91.1% lipids (w/w) (ratio 0.9:9.1). Repeated extrusions are performed using a Thermobarrel Extruder (Northern Lipids Inc., Vancouver BC) through membranes of 0.2, 0.1, 0.08 and 0.05 μm pore sizes (Whatman) under pressure in ultra pure nitrogen. About 15 passages were used and the average particle diameter and size distribution at a 90° angle were controlled with dynamic light scattering (N4+ nanoparticle analyzer, Beckman-Coulter). The total lipid to cisplatin ratio in Lipoplatin is 10.24:1 (w/w) mg lipid/mg cisplatin. The content of Lipoplatin in cholesterol is 11.6% (w/w) of the total lipids. It can been calculated that 1.3 mg cholesterol are injected per mg of cisplatin in its Lipoplatin formulation.

Of an average size of 110 nm, the type of liposome particles used in Lipoplatin is a proprietary formulation. Lipoplatin is provided in 50-ml clear glass vials of 3 mg/ml (concentration refers to cisplatin). The concentration of 3 mg/ml of cisplatin in Lipoplatin exceeds the solubility of the free drug, cisplatin, usually provided as a 0.5 -1 mg/ml solution for IV infusion. Although cisplatin is light-sensitive, the Lipoplatin formulation appears to be light resistant presumably because liposomes shield the drug. However, as a precaution, storage in a dark area is recommended. Lipoplatin is stored at 4°C and has an expiration date of two years.

Study design. Patient selection criteria included: histologically confirmed malignancy type, stage IV disease, a life expectancy of > 3 months, patients had previously undergone standard treatment relevant to the kind of malignancy, a performance status of 0-2 and age > 18. All patients gave their signed informed consent to participate in the study. Other eligibility criteria included adequate bone marrow function (peripheral absolute neutrophil count ≥ 2,000 /mm^3, platelet count > 100,000/mm^3, and haemoglobin > 9 g/dl), adequate liver function (total bilirubin ≤ 1.5 mg/dl, SGOT, or SGPT no greater than 4 times normal) and adequate renal function (creatinine < 1.5 mg/dl). In patients with nephrectomy creatinine clearance was performed. The protocol was approved by the Hellenic Drug Organization.

Lipoplatin was given as an 8 h i.v. infusion; 8 hours was chosen in order to be able to control possible adverse effects occurring during drug infusion since this study is the first report on the treatment of human subjects with Lipoplatin.

All 27 patients were at stage IV (19 pancreatic carcinoma, 6 renal cell carcinoma, 1 with gastric cancer and 1 with squamous cell carcinoma of the head and neck). All had a histologically confirmed diagnosis. In all cases, Lipoplatin was a second- or third-line treatment and was administered when the disease was refractory to standard treatment.

Before study entry, all patients underwent the following: physical examination, tumor measurement or evaluation, WHO performance status, ECG, full blood, liver and renal function tests urinalysis. Staging was determined by chest and abdominal CT scans, bone scans and occasional magnetic resonance imaging. The patients' characteristics are shown in Table I. Full blood count (FBC), liver function tests, plasma protein, blood urea, and plasma creatinine preceded every course of treatment.

Table 1. Patients' characteristics.

Gender	
Male	16
Female	11
Age (years)	
Median	62
Range	39-78
Histology	
Adenocarcinoma	26
Pancreatic	19
Renal	6
Gastric	1
Squamous cell carcinoma (head and neck)	1
Prior Treatment	
Pancreatic cancer	gemcitabine + irinotecan
Renal cell cancer	IFN-α
	vinblastine
gastric cancer	leucovorin + 5 FU + etoposide
SCC head and neck	cisplatin + 5-FU
	carboplatin + paclitaxel
WHO performance status	
0	1
1	16
2	10

Treatment. Lipoplatin was administered as an 8 h infusion diluted in 1 L 5% dextrose, repeated every two weeks. FBC was repeated one week after each cycle. The total number of cycles was 92 (median 4, range 2-5).

Lipoplatin 25 mg/m^2 was administered to 3 patients, 50 mg/m^2 to 3, 75 mg/m^2 to 4, 100 mg/m^2 to 12 and 125 mg/m^2 to 5 patients. Three additional patients were each treated once at 200, 250 and 300 mg/m^2.

Chemical evaluation, full blood count, blood urea, plasma creatinine and liver function tests were done before treatment and one week after. In 6 nephrectomized patients creatinine clearance was performed before and after the end of the treatment courses. Lipoplatin dosage escalation is shown in Table II.

Patient evaluation. For all patients, complete medical history, complete physical examination, an electrocardiogram and a neurological examination were done before treatment. Prior to each Lipoplatin administration and 7 days after, complete haematological analysis, plasma chemistry including cholestero

Table. II Lipoplatin dosage escalation

Dose Level (mg/m$^{2)}$	Patients (no.)
25	3
50	3
75	4
100	12
125	5
Total no. of patients	27

and triglyceride levels, an electrocardiogram, urine analysis and neurological examination were performed. Measurable tumor lesions were evaluated before and after two to three cycles. Toxicity was graded according to World Health Organization (WHO) criteria and dose-limiting toxicities (DLTs) were defined as grade 3 and 4 myelotoxicity and gastrointestinal toxicity, with the exception of nephrotoxicity, ototoxicity, alopecia, weight change, and fatigue which were not considered dose-limiting. Had there been plasma creatinine increase, then creatinine clearance would have been performed.

Analysis of plasma ultrafiltrates for platinum. For pharmacokinetic studies, blood was drawn at 0, 3, 6, 8, 12, 24 h and 3, 5, 7 days into tubes containing EDTA and total platinum levels (i.e. free plus protein-bound plus liposomal) were analyzed by atomic absorption. Blood was collected from patients and centrifuged to remove cells. Total plasma was analyzed for platinum. Total platinum was also determined in the ultrafiltrate of a smaller number of plasma as follows: approximately 1 ml of plasma was placed in an Amicon ultracentrifugal filter device with a 10,000 molecular weight cut-off (Millipore, USA). Tubes (filter devices) were centrifuged at 4°C at 2,500 g for 10 min in a Sorval centrifuge (swinging bucket rotor) and platinum levels were measured separately in the ultrafiltrate. Total platinum concentrations were determined in diluted plasma samples and free platinum was determined in the ultrafiltrate samples. Urine was also collected and total platinum concentration and total levels were estimated by atomic absorption.

Total plasma was analyzed in an Atomic absorption spectrophotometer using both the Perkin Elmer Analyst 200 (flame) and the Perkin Elmer AA 700 Graphite Furnace Atomic Absorption Spectrometers.

Pharmacokinetic parameters. Blood samples were collected into tubes containing EDTA and placed on an ice-bath immediately after withdrawal. Whole blood was centrifuged at 4°C for 5 min (3,000 g). Aliquots (0.1 ml) of the obtained plasma fraction were diluted tenfold (1:9) with water and were further diluted by the autosampler function of the Atomic Absorption instrument (AA700).

Non-compartmental methods were applied to calculate the pharmacokinetic parameters of total platinum. The area under the plasma concentration-time curve (AUC) was determined using the linear trapezoidal method with extrapolation to

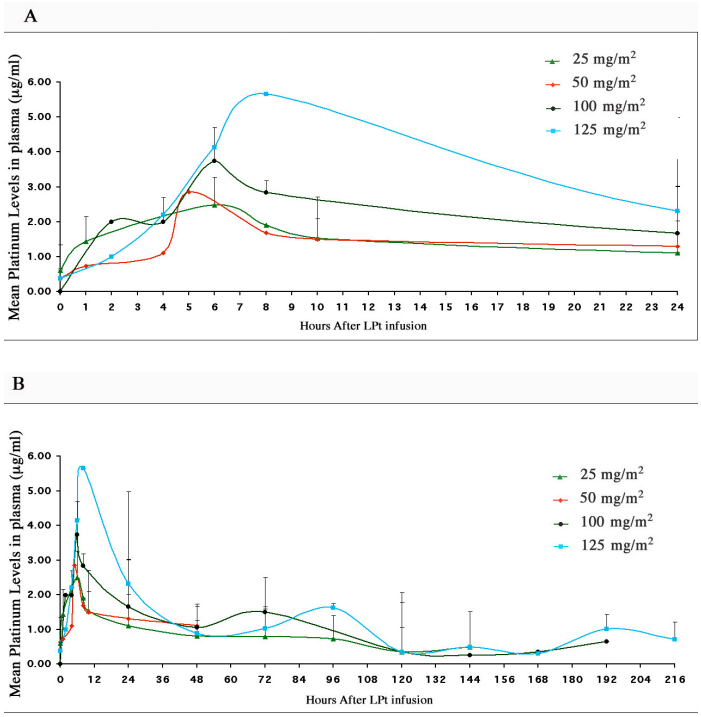

Figure 1. (A), Depiction of mean plasma concentration-time curves per dose level. Platinum levels in µg/ml in plasma of patients as a function of time (up to 7 days) are shown. The data are based on 17 patients with stage IV advanced cancers; each patient received an average of 2.5 infusions at 25, 50, 75, 100, 125 mg/m^2. (B), Same curve up to 7 days.

infinity. The elimination rate constant for total platinum (k) was calculated by linear regression analysis of the logarithmic plasma concentration-time curve. Total body clearance (Cl), the elimination half-life ($t_{1/2}$) and the volume of distribution at steady state (V_{ss}) were calculated using standard equations (17).

Results

Pharmacokinetics of Lipoplatin. Total platinum concentrations were measured in the diluted plasma samples and free plasma concentrations were measured in the plasma ultrafiltrates. Complete plasma concentration-time curves for total platinum were obtained in 17 patients out of the 27 and used for pharmacokinetic analyses. The mean plasma concentration-time curves per dose level are shown in Fig. 1. During the 8 h period of Lipoplatin infusion and for 7 days thereafter, the concentration of total platinum was measured using atomic absorption spectrometry. During the 8 h infusion period, the maximum platinum level in the plasma is attained at 6 h at doses of 25, 50, 75, and 100 mg/m^2 and declines thereafter. The infusion of Lipoplatin at 125 mg/m^2 rendered maximal levels of platinum in the plasma at 8 h.

Differences in plasma levels of platinum at different administered doses were registered: after an infusion of 25 mg/m^2, a maximum of 2.5 µg/ml was attained in the plasma, at 50 mg/m^2 2.9 µg/ml (at 6h), at 100 mg/m^2 3.7 µg/ml (at 6h) and at 125 mg/m^2 the maximum attained in the plasma was 5.7 µg/ml (at 8h). The levels of platinum in the blood after Lipoplatin infusion drop to normal on the fourth day at a

Table. III. Pharmacokinetic parameters of total platinum in patients' plasma at the different dose levels.

Dose mg/m^2	Pts (n)	AUC (hΞµg/ml)	C_{max} (µg/ml)	Cl (L/m^{2}h)	K_{el} (1/h)	$t_{1/2}$ (h)	V_{ss} (L/m^2)
25	5	139.63	2.48±1.18	0.18	0.0114	60.79	15.71
50	3	119.19	2.87±0.59	0.42	0.0001	N/A	N/A
100	5	172.89	3.74±1.18	0.58	0.0059	117.46	98.03
125	4	256.09	5.65±2.67	0.49	0.0085	81.53	57.42

N/A non applicable

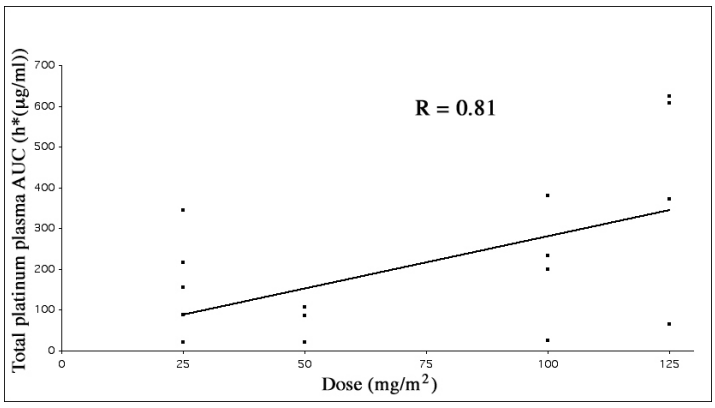

Figure 2. Plot of the total platinum AUC versus the administered dose of Lipoplatin.

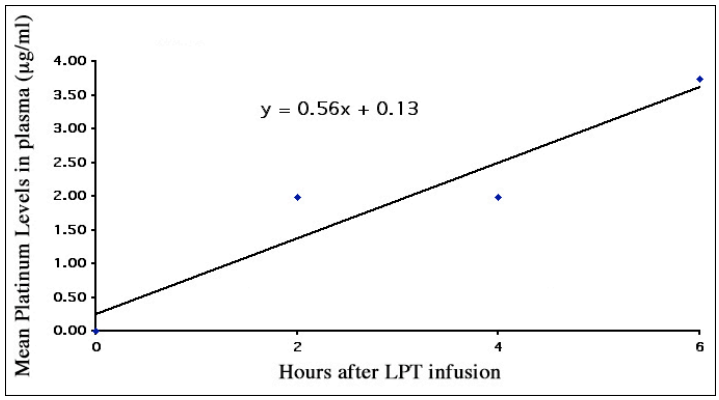

Figure 3. Formula depicting the mean platinum levels in the plasma during the first 6 h of the infusion at a dose of 100 mg/m^2.

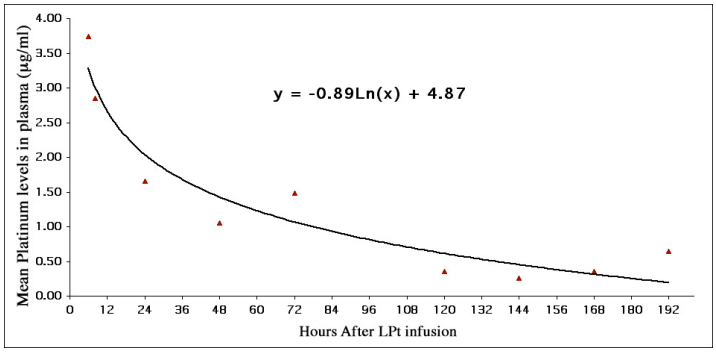

Figure 4. Formula depicting the mean platinum levels in the plasma after the 6 h infusion at a dose of 100 mg/m^2.

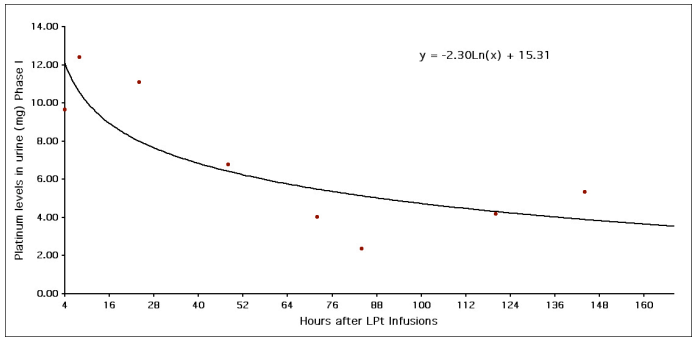

Figure 5. Formula depicting the mean levels of platinum excreted in the urine (total mg) as a function of time (in hours) after a dose of 100 mg/m^2 Lipoplatin in cancer patients.

dose of 100 mg/m^2, but at a dose of 125 mg/m^2 platinum can be detected in the blood for 7 days (Fig. 1).

Mean pharmacokinetic parameters for total platinum calculated for the 25, 50, 100 and 125 mg/m^2 doses are shown in Table 3. At the 100 mg/m^2 dose, the AUC, determined using the linear trapezoidal method with extrapolation to infinity (17), was 172.89 µgΞh/ml.

The maximum concentration of total platinum in plasma reached (C_{max}) was 3.74±1.18 µg/ml. Total body clearance (Cl) was 0.58 L/(m^{2}Ξh). This was calculated by Cl=$D_{i.v.}$/AUC, where $D_{i.v.}$ is the intravenous dose of Lipoplatin and AUC the relative area under the curve for this specific dose.

The elimination rate constant (K_{el}) was 0.0059 h^{-1}. This was calculated by linear regression analysis of the logarithmic plasma concentration-time curve by the formula K_{el} = [Ln(C_{p1})-Ln(C_{p2})]/(t_2-t_1) where t_1 and t_2 are the starting and ending time points of measurements and C_{p1} and C_{p2} the starting and ending concentrations of total platinum in plasma for t_1 and t_2, respectively.

The elimination half-life ($t_{1/2}$) was 117.46 h. This was calculated by the formula: $t_{1/2}$= 0.693 (1/K_{el}). 1/K_{el} is the mean residence time (MRT), the statistical moment analogy to half-life $t_{1/2}$ (17).

Table IV. Adverse reactions following Lipoplatin infusion.

	Patients (%)			
	Grade 1	Grade 2	Grade 3	Grade 4
Neutropenia	14.81	3.7	-	-
Thrombocytopenia	-	-	-	-
Anemia (hemoglobin)	19	-	-	-
Nausea/vomiting	7.5	-	-	-

Liver function tests (SGOT (serum glutamic oxaloacetic transaminase) SGPT, γ-GT, alkaline phosphatase, bilirubin) were within normal limits before and after treatment. Renal function tests (blood urea, serum creatinine and creatinine clearance) showed no change before and after treatment. Grade 1 and 2 toxicity was only observed at the dose of 125 mg/m^2. At the start of the infusion, 8/27 patients suffered from acute severe epigastric and back pain which lasted for about 5 minutes and then subsided spontaneously without the administration of analgesics. No cardiotoxicity, ototoxicity or neurotoxicities were observed, and there was no allergic reaction or hair loss.

The volume of distribution at steady state (V_{ss}) was 98.03 L/m^2. This was calculated by the formula: V_{ss} = Cl (1/K_{el}). A plot of the total platinum plasma AUC versus the administered dose of Lipoplatin is shown in Fig. 2 where the solid line is the regression line (R = 0.81).

The curve shown in Fig. 1 was divided into two parts (from 1 to 6 h in Figure 3 and from 6h to 172 h from infusion in Figure 4) in order to derive two equations to describe the distribution of platinum levels in plasma as a function of time. The kinetics of platinum increase in plasma as a function of time can be described during the initial 6 h by the formula y = 0.56x + 0.13 where y is the platinum level in plasma in µg/ml and x is the time in hours from the start of the infusion. From 6 h and thereafter the levels of platinum constantly decline. The kinetics of platinum from 6 h to 3 days for the dose of 100 mg/m^2 can be described by the formula: y = -0.89Ln(x) + 4.87.

Excretion of Lipoplatin in the urine. Excretion of platinum in the urine in Lipoplatin-treated patients attains a maximum within 8 h (infusion period) and declines thereafter.

When a dose of 100 mg/m^2 was infused in a patient (1.6 m^2 body surface area) 9.1% was excreted in the urine during the 8 h of i.v. infusion, 16.8% during the following 16 h and 10% during the following 24 h. During the third day, an additional 4.8% was excreted in the urine. Therefore, during the 3 days from the start of the Lipoplatin infusion about 40.7% of total platinum is excreted in the urine. During the following days the amount excreted dropped.

The kinetics of platinum excretion in the urine after the 8 h infusion period can be described by the formula: y = -2.3 Ln(x) + 15.3 where y is the total platinum in mg and x is the time from the start of the infusion in hours (Fig. 5).

Levels of platinum in ascites fluid. The levels of platinum in ascites fluid after Lipoplatin infusion in two patients showed levels of 0.52 μg/ml at 8h, 1.08 μg/ml at 24h which declined thereafter to 0.54 μg/ml at 48h, and 0.34 μg/ml at 6 days (average from 2 patients).

Toxicity of Lipoplatin. Grades 1 and 2 myelotoxicity (neutropenia) and grades 1 and 2 GI tract toxicity (vomiting) were observed only at the dose of 125 mg/m^2. No other toxicity was observed even with repeated doses. At the beginning of the infusion 8/27 patients (29.6%) described acute severe epigastric and back pain which lasted for about 5 min and subsided spontaneously without analgesic administration. Overall, treatments in repeated doses every two weeks were well-tolerated. It is interesting to note that three patients with mild renal insufficiency and with plasma creatinine of 1.5-2.2, treated with a dose of 100 mg/m^2, showed no increase in plasma creatinine. No change in creatinine clearance was observed comparing the values before and after treatment in three out of six patients who had creatinine clearance 35-50 ml per minute (the normal limits of creatinine clearance are 60 ml/min; this limit varies from 60-70 ml/min according to the body surface area). No aggravation of hematological toxicity was observed. Adverse reactions are shown in detail in Table IV.

Response. Although response measurement was not a primary goal of the study, 3/27 (11.1%) patients achieved partial response; of the remaining 24 patients, 14 (51.9%) achieved stable disease and clinical benefit. Follow up was 2-5 months.

Discussion

The clinical use of a new liposomal formulation of cisplatin is described in this study for the first time in the literature. The Lipoplatin formulation uses several advancements in its liposome encapsulation: i) the anionic lipid DPPG gives Lipoplatin its fusogenic properties presumably acting at the level of entry of the drug through the cell membrane after reaching the target tissue; ii) the total lipid to cisplatin ratio is low (10.24:1 mg lipid/mg cisplatin) in Lipoplatin which means that less lipid is injected into the patient. By comparison, the ratio of lipids to cisplatin in the liposomal formulation SPI-77 is 71.43:1 (18) which is 7-fold higher lipids per mg cisplatin compared to Lipoplatin; and iii) The PEG polymer coating used on Lipoplatin is meant to give the drug particles the ability to pass undetected by the macrophages and immune cells, to remain in circulation in body fluids for long periods and tissues and to extravasate preferentially and infiltrate solid tumors and metastases through the altered and often compromised tumor vasculature.

Liposomal cisplatin is a new cisplatin modification, which administered at 125 mg/m^2 has no renal toxicity (Table IV). Animal studies by other investigators have shown that cisplatin has caused damage to the proximal kidney tubules because the platinum-sulfhydryl group complexes which form, are taken up by the kidney cells through an organic anion transport mechanism. Pretreatment of the animals with the glutathione synthesis inhibitor, buthionine sulfoximine, potentiated the tubule damage; the stability of these complexes was found to be dependent on the intracellular glutathione (GSH) level, and they were gradually metabolized to reactive metabolite(s) by renal intracellular beta-lyase and S-oxidase (19). Cisplatin accumulates in cells from all nephron segments but is preferentially taken up by the highly susceptible proximal tubule cells within the S3 segment, which bear the brunt of the damage (20,21).

Nephrotoxicity following cisplatin treatment is common and may manifest after a single dose with acute renal failure or may present with a chronic syndrome of renal electrolyte wasting. Despite various hydration protocols designed to minimize the nephrotoxicity, approximately one-third of patients who receive cisplatin develop evidence for acute renal failure (22).

We believe that Lipoplatin does not cause the extensive damage to the proximal kidney tubules that non-liposomal cisplatin does for the following reasons: 1. The reactivity of cisplatin in its Lipoplatin formulation is extremely hindered because of the protection offered by the lipid capsule; release of cisplatin from the liposome (for example via fusion with the cell membrane) is expected to render the drug active inside the cell where its cytotoxic effects are needed. It is proposed that entrance of Lipoplatin particle inside the kidney tubule cells is limited. 2. Lipoplatin is released through the kidney with a half-life of 60-117 h compared to 6.5 h for cisplatin. 3. One additional reason for the lower nephrotoxicity of Lipoplatin arises from the lower levels of total platinum in kidneys in animal studies after Lipoplatin compared to cisplatin treatments. Indeed, the maximum level of total platinum in rat kidneys after intraperitoneal bolus injection of cisplatin or Lipoplatin at similar doses were similar (attained in 15 min), but after approximately 1h from intraperitoneal injection the steady state accumulation of total platinum in kidney was 5 times higher for cisplatin compared to Lipoplatin. In practical terms this means that the uptake of total platinum in kidneys after Lipoplatin is 5 times lower that after cisplatin chemotherapy. These pharmacokinetic differences may account, at least in part, for the low renal toxicity of Lipoplatin which was documented in animals using tubule cell necrosis and apoptosis, as well impaired renal function assays (16).

With cisplatin, at a dose range of 40-140 mg/m^2 given as a bolus injection or as 1-24h infusions, 10-40% of the platinum is excreted in the urine in 24h; plasma concentrations of cisplatin decay with a half-life of 20-30 min following bolus administrations of 50-100 mg cisplatin/m^2 (23-25). The release of Lipoplatin in the urine was much slower compared to cisplatin. We suggest that with the Lipoplatin infusion there may be an initial compartmentalization into body tissues which is followed by release into body fluids at a later time.

Lipoplatin is different from the SPI-77 liposomal formulation of cisplatin (26,27) in pharmacokinetics. SPI-77 has a half-life of 134 h and urine excretion reaching only 4% of the total dose in 72 h (18).

Other liposomally-encapsulated drugs such as the liposomal doxorubicin known as Doxil (Caelyx in Europe) have been extensively used in the clinic. Doxil is unable to cross the cell membrane barrier and thus, doxorubicin is released into the extracellular space after digestion of the lipid capsule by extracellular lipases over periods of days (28).

The pharmacokinetics of this Phase I study comparing plasma levels and urine excretion show a slow release of platinum in the plasma, which gradually drops over a period of 3 days. We have deduced that the slow excretion and release of platinum results in low nephrotoxicity. It is worth mentioning that the SPI-77 formulation of cisplatin showed a total body clearance of 14-30 ml/h which was significantly lower than that reported for cisplatin (20 L/m^2 per h due to the slow release of cisplatin from liposomes) (29). Similarly, Lipoplatin showed a total body clearance of 0.18 L/(m^2Ξh) (at the dose of 25 mg/m^2) and 0.49 L/(m^2Ξh) (at the dose of 125 mg/m^2) (Table III) which is also considerably lower than the total body clearance for cisplatin.

We did not reach the MTD even when we increased the dose up to 350 mg/m^2 in one patient as a single infusion but we did observe the first signs of toxicity. Since we went as high as double the dose of cisplatin and as the future plan would be the combination of Lipoplatin with other cytotoxic drugs we ended the experimental trial at this point. The toxicity of cytotoxic agents is modified and aggravated when these agents are administered in combination. It is worth mentioning that the dose of cisplatin as well as of other cytotoxic drugs used in combination in clinical practice is always considerably below the MTD.

In conclusion, the results of this Phase I study have shown that the pharmacokinetic behavior of the total platinum in the Lipoplatin formulation is significantly altered with respect to free cisplatin. The pharmacokinetics of Lipoplatin are mainly dominated by its liposomal properties.

The repetition of 5 cycles every two weeks at a dose of Lipoplatin 100 mg/m^2 showed no cumulative adverse reaction with respect to renal, neuro-, gastrointestinal or myelo-toxicity.

References

1. Rosenberg B: Noble metal complexes in cancer chemotherapy. Adv Exp Med Biol 91: 129-50, 1977.
2. Livingston RB: Combination chemotherapy of bronchogenic carcinoma. I. Non-oat cell. Cancer Treat Rev 4:153-65, 1977.
3. Al-Sarraf M: Chemotherapeutic management of head and neck cancer. Cancer Metastasis Rev 6: 181-98, 1987.
4. Sternberg CN, Yagoda A, Scher HI, Watson RC, Geller N, Herr HW, Morse MJ, Sogani PC, Vaughan ED, Bander N, et al.: Methotrexate, vinblastine, doxorubicin, and cisplatin for advanced transitional cell carcinoma of the urothelium. Efficacy and patterns of response and relapse. Cancer 64: 2448-58, 1989.
5. Oliver T and Mead G: Testicular cancer. Curr Opin Oncol 5: 559-567, 1993.
6. Taylor AE, Wiltshaw E, Gore ME, Fryatt I, Fisher C: Long-term follow-up of the first randomized study of cisplatin versus carboplatin for advanced epithelial ovarian cancer. J Clin Oncol 12: 2066-70, 1994.
7. Non-small Cell Lung Cancer Collaborative Group: Chemotherapy in non-small cell lung cancer: a meta-analysis using updated data on individual patients from 52 randomised clinical trials. BMJ 311: 899-909, 1995
8. Caraceni A, Martini C, Spatti G, Thomas A, Onofrj M (1997) Recovering optic neuritis during systemic cisplatin and carboplatin chemotherapy. Acta Neurol Scand 96: 260-261
9. Stathopoulos GP, Rigatos S, Malamos NA (1999) Paclitaxel combined with cis-platin as second-line treatment in patients with advanced non-small cell lung cancers refractory to cis-platin. Oncol Rep 6: 797-800
10. Piccart MJ, Bertelsen K, James K, Cassidy J, Mangioni C, Simonsen E, Stuart G, Kaye S, Vergote I, Blom R, Grimshaw R, Atkinson RJ, Swenerton KD, Trope C, Nardi M, Kaern J, Tumolo S, Timmers P, Roy JA, Lhoas F, Lindvall B, Bacon M, Birt A, Andersen JE, Zee B, Paul J, Baron B, Pecorelli S (2000) Randomized intergroup trial of cisplatin-paclitaxel versus cisplatin-cyclophosphamide in women with advanced epithelial ovarian cancer: three-year results. J Natl Cancer Inst 92: 699-708
11. Boulikas T and Vougiouka M (2003) Cisplatin and platinum drugs at the molecular level. Oncol Rep 10: 1663-1682
12. Boulikas T and Vougiouka M: Recent clinical trials using cisplatin, carboplatin and their combination chemotherapy drugs (review). Oncol Rep. 11: 559-595, 2004.
13. Humes HD: Insights into ototoxicity. Analogies to nephrotoxicity. Ann NY Acad Sci 884: 15-18, 1999
14. Arany I, Safirstein RL: Cisplatin nephrotoxicity. Semin Nephrol 23: 460-4, 2003.
15. Boulikas T: Low toxicity and anticancer activity of a novel liposomal cisplatin (Lipoplatin) in mouse xenografts. Oncol Rep 12: 3-12, 2004.
16. Devarajan P, Tarabishi R, Mishra J, Ma Q, Kourvetaris A, Vougiouka M, and Boulikas T: Low Renal Toxicity Of Lipoplatin Compared To Cisplatin In Animals. Anticancer Research 24: 2193-2200, 2004.
17. Gibaldi M, Perrier D: Noncompartmental analysis based on the statistical moment theory. In Pharmacokinetics, Gibaldi M, Perrier D (eds), pp 409-417, 2nd edn. Marcel Dekker: New York, 1982.
18. Veal GJ, Griffin MJ, Price E, Parry A, Dick GS, Little MA, Yule SM, Morland B, Estlin EJ, Hale JP, Pearson AD, Welbank H, Boddy AV: A phase I study in paediatric patients to evaluate the safety and pharmacokinetics of SPI-77, a liposome encapsulated formulation of cisplatin. Br J Cancer 84: 1029-35, 2001.
19. Ban M, Hettich D, Huguet N: Nephrotoxicity mechanism of cis-platinum (II) diamine dichloride in mice. Toxicol Lett 71: 161-8, 1994.
20. Park MS, De Leon M, Devarajan P: Cisplatin induces apoptosis in LLC-PK1 cells via activation of mitochondrial pathways. J Am Soc Nephrol 13: 858-865, 2002.
21. Hanigan MH, Devarajan P: Cisplatin nephrotoxicity: molecular mechanisms. Cancer Therapy 1: 47-61, 2003.
22. Santoso JT, Lucci JA 3rd, Coleman RL, Schafer I, Hannigan EV.: Saline, mannitol, and furosemide hydration in acute cisplatin nephrotoxicity: a randomized trial. Cancer Chemother Pharmacol 52: 13-8, 2003.
23. Gullo JJ, Litterst CL, Maguire PJ, Sikic BI, Hoth DF, Woolley PV.: Pharmacokinetics and protein binding of cis-dichlorodiammine platinum (II) administered as a one hour or as a twenty hour infusion. Cancer Chemother Pharmacol 5: 21-6, 1980.
24. Ostrow S, Egorin M, Aisner J, Bachur N, Wiernik PH.: High-dose cis-diamminedichloro-platinum therapy in patients with advanced breast cancer: pharmacokinetics, toxicity, and therapeutic efficacy. Cancer Clin Trials 3: 23-7, 1980.
25. Pratt CB, Hayes A, Green AA, Evans WE, Senzer N, Howarth CB, Ransom JL, Crom W.: Pharmacokinetic evaluation of cisplatin in children with malignant solid tumors: a phase II study. Cancer Treat Rep 65:1021-6, 1981.
26. Bandak S, Goren D, Horowitz A, Tzemach D, Gabizon A: Pharmacological studies of cisplatin encapsulated in long-circulating liposomes in mouse tumor models. Anticancer Drugs 10: 911-20, 1999.
27. Newman MS, Colbern GT, Working PK, Engbers C, Amantea MA: Comparative pharmacokinetics, tissue distribution, and therapeutic effectiveness of cisplatin encapsulated in long-circulating, pegylated

liposomes (SPI-077) in tumor-bearing mice. Cancer Chemother Pharmacol 43: 1-7, 1999.

28. Martin F and Boulikas T: The challenge of liposomes in gene therapy. Gene Ther Mol Biol 1: 173-214, 1998.

29. Meerum Terwogt JM, Groenewegen G, Pluim D, Maliepaard M, Tibben MM, Huisman A, ten Bokkel Huinink WW, Schot M, Welbank H, Voest EE, Beijnen JH, Schellens JM: Phase I and pharmacokinetic study of SPI-77, a liposomal encapsulated dosage form of cisplatin. Cancer Chemother Pharmacol 49: 201-10, 2002