Eur J Clin Pharmacol (2001) 57: 393–402
DOI 10.1007/s002280100319

PHARMACOKINETICS AND DISPOSITION

**Bernhard Erdlenbruch · Marit Nier · Wolfgang Kern
Wolfgang Hiddemann · Arnulf Pekrun · Max Lakomek**

Pharmacokinetics of cisplatin and relation to nephrotoxicity in paediatric patients

Received: 15 November 2000 / Accepted in revised form: 2 May 2001 / Published online: 16 June 2001

Abstract *Background*: Cisplatin is a highly effective and frequently used drug in the chemotherapy of solid tumours in children, but only limited data are available on the pharmacokinetics of cisplatin and its associated nephrotoxicity in paediatric patients.
Methods: We investigated the pharmacokinetics of free platinum (Pt) in 12 children (25 courses) receiving cisplatin (75–120 mg/m^2) either as a continuous 72-h infusion, prolonged single 6-h infusion or repetitive 1-h infusions. Plasma and urinary Pt concentrations were analysed using atomic absorption spectroscopy. Cisplatin-induced nephrotoxicity was determined using creatinine clearance and several glomerular and tubular marker proteins.
Results: Using a two-compartment model the pharmacokinetic parameters for free Pt were: initial half-life 21.6 ± 9.6 min, terminal half-life 25.9 ± 16.2 h, area under the plasma concentration–time curve (AUC) 13.5 ± 4.97 (μg/ml)·h/(100 mg/m^2) and cumulative renal elimination$_{\text{infinity}}$ 41.7 ± 6.6% of dose. Higher cisplatin delivery rates led to higher peak concentrations of free Pt in plasma and urine and to lower cumulative renal Pt elimination ($P < 0.01$). During all courses, increases of urinary albumin and α1-microglobulin excretion were documented. The creatinine clearance decreased significantly to 70% of baseline values. Correlations were found between both peak free Pt concentrations in plasma and in urine and the maximum of urinary excretions of albumin and of *N*-acetyl-β-D-glucosaminidase and the nadir of the glomerular filtration rate ($P < 0.05$).
Conclusions: With respect to nephrotoxicity, long-term infusions of cisplatin seem to be preferable over intermittent bolus administration in paediatric patients. The best predictive pharmacokinetic parameters for cisplatin-associated nephrotoxicity in children are peak free Pt concentrations in plasma and urine.

Keywords Cisplatin pharmacokinetics · Nephrotoxicity · Children

B. Erdlenbruch (✉) · M. Nier · A. Pekrun · M. Lakomek
Kinderklinik der Universität Göttingen,
Robert-Koch-Strasse 40, 37075 Göttingen, Germany
E-mail: erdlenbr@med.uni-goettingen.de
Tel.: +49-551-396210
Fax: +49-551-396231

W. Kern · W. Hiddemann
Medizinische Klinik und Poliklinik III der Universität München,
Marchioninistrasse 15, 81366 München, Germany

Introduction

Cisplatin (CDDP) is a potent anticancer drug and well established in the chemotherapy of a variety of solid tumours in paediatric patients, including osteosarcoma, neuroblastoma [1], medulloblastoma and germ cell tumours [2]. However, administration of CDDP is associated with myelosuppression, gastrointestinal toxicity, peripheral neuropathy, ototoxicity and nephrotoxicity [3, 4], which is the dose-limiting factor. Due to the favourable treatment outcomes of the majority of children with cancer, long-term sequelae as a result of highly organotoxic antineoplastic treatment are becoming more and more important. Both acute nephrotoxicity and chronic renal damage are well documented after CDDP administration [5, 6] in adults as well as in paediatric patients [7]. Several strategies have been developed in order to prevent CDDP-induced nephrotoxicity, such as forced diuresis by extensive fluid delivery and mannitol treatment, saline loading using 3% saline as a vehicle for CDDP [8] or concomitant administration of sodium thiosulfate [9, 10]. Despite these clinical interventions, only partial protection of the kidneys can be achieved and severe nephrotoxicity may occur after administration of therapeutic doses. However, there is evidence that CDDP pharmacokinetics influence the platinum (Pt)-induced renal damage [11], but only a few investigations have been performed in adults to clarify this issue. Moreover, despite its widespread use in the chemotherapy of paediatric solid tumours, very limited

data on the pharmacokinetics of CDDP in children are available and most of our knowledge about CDDP pharmacokinetics stems from studies in adults.

High plasma or urinary Pt levels are thought to increase the incidence of CDDP-related nephrotoxicity [12, 13, 14]. Therefore, fractional bolus injections or continuous intravenous (i.v.) infusions have become standard protocols for cancer treatment. In clinical studies, it was demonstrated that prolonged dosing schedules of CDDP are as effective as bolus injections and are associated with less gastrointestinal and renal toxicity [15, 16, 17]. Pharmacokinetic evaluations revealed significantly lower peak plasma Pt concentrations with 5-day continuous infusions than intermittent fractional bolus injections [18, 19, 20]. Divergent results, however, were reported concerning the area under the plasma concentration–time curve (AUC) of free Pt. Forastiere [19] and co-workers described a twofold higher AUC in patients treated by prolonged infusion periods whereas no differences were found by other groups [18, 20]. In conclusion, no consistent reports exist so far on correlations between CDDP pharmacokinetics and CDDP-induced nephrotoxicity in patients.

The few studies of CDDP pharmacokinetics carried out in children provide only limited information. This is in part due to the small number of patients observed [21, 22], but also due to differences between the infusion schedules investigated. Peng and co-workers [23] provided a broad spectrum of pharmacokinetic data in children receiving a 24-h infusion of 50–120 mg/m^2 while Crom et al. [24] described the total Pt disposition in children after a 6-h infusion of 30 mg/m^2 or 90 mg/m^2. The results of both studies were similar to those found in adults. Dominici et al. [25] evaluated pharmacokinetic parameters of high-dose CDDP, which do not necessarily apply to standard therapeutic regimens. In children, no attempt has yet been made to demonstrate the relationship between CDDP pharmacokinetics and its nephrotoxic effects. Therefore, the objectives of the present study were to describe CDDP pharmacokinetics in children when given as standard combination therapy and to identify correlations between pharmacokinetic parameters and Pt-associated nephrotoxicity.

Materials and methods

Patients and drug administration

The study subjects were 12 children and adolescents hospitalised in the department of paediatrics of the University of Göttingen due to different malignant solid tumours. Age varied from 5 years to 19 years (mean 11 years; five males and seven females). All patients received CDDP chemotherapy in combination with other cytostatic drugs according to the respective treatment protocols. Due to insufficient co-operation of the patients, loss of samples or technical reasons not all cisplatin courses administered to the patients could be used for further investigation. A total of 25 courses (1–4 courses per patient) was included in the study. All children and their parents gave informed consent prior to entering the study in accordance with the requirements of the university ethics committee. Characteristics of the children are summarised in Table 1. All patients had normal renal and hepatic function before entering the study. Cisplatin (Platinex) was administered in physiological saline either as repeated 1-h infusions over a period of three (medullo-

Table 1 Characteristics of patients and infusion schedules for cisplatin (CDDP) treatment. Standard treatment protocols of the German Society of Paediatric Haematology and Oncology (GPOH) were used. *LC* lymphoepithelial carcinoma, *OS* osteosarcoma, *MB* medulloblastoma, *DG* dysgerminoma, *5-FU* 5-fluorouracil, *MTX* methotrexate, *ADM* adriamycin, *IFO* ifosfamide, *CA* cytarabine, *VP-16* etoposide, *BLM* bleomycin, *HIT* brain tumour study, *COSS* co-operative osteosarcoma study, *MAKEI* malignant germ cell tumour study

Patient	Age (years)	Gender	Weight (kg)	Surface area (m^2)	Diagnosis	Name of protocol	Dose per cycle (mg/m^2)	Infusion time (h)	Cycles studied (*n*)	Other medication
1	16	Male	55.2	1.67	LC	LC	75	6	3	5-FU, MTX
2	14	Female	58	1.63	OS	COSS-86c	120	72	3	MTX, ADM, IFO, VP-16
3	6	Female	21	0.85	OS	COSS-86c	120	72	2	MTX, ADM, IFO, VP-16
4	18	Female	67.9	1.8	OS	COSS-86c	120	72	1	MTX, ADM, IFO, VP-16
5	9	Female	26.7	1.0	OS	COSS-86c	120	72	2	MTX, ADM, IFO, VP-16
6	10	Female	38.3	1.2	MB	HIT-91	3×40	3×1	2	MTX, CA, IFO, VP-16
7	8	Male	28.7	1.03	MB	HIT-91	3×40	3×1	1	MTX, CA, IFO, VP-16
8	14	Female	54.8	1.56	MB	HIT-91	3×40	3×1	2	MTX, CA, IFO, VP-16
9	7	Male	18.2	0.78	MB	HIT-91	3×40	3×1	2	MTX, CA, IFO, VP-16
10	5	Male	19.9	0.81	MB	HIT-91	3×40	3×1	2	MTX, CA, IFO, VP-16
11	6	Male	22.5	0.88	MB	HIT-91	3×40	3×1	1	MTX, CA, IFO, VP-16
12	19	Female	44.5	1.44	DG	MAKEI-89	5×20[a]	5×1	4	BLM, VP-16

[a]Cisplatin dose in the first course amounted to only 5×15 mg/m^2

blastoma, 3×40 mg/m^2) or five consecutive days (dysgerminoma, 5×20 mg/m^2), as a prolonged single infusion over 6 h (lymphoepithelial carcinoma, 75 mg/m^2) or as a continuous i.v. infusion over 72 h (osteosarcoma, 120 mg/m^2). Nine patients received several CDDP courses during the study (six patients got two courses, two got three courses and one underwent four courses). For hydration, a continuous infusion of 3 l/m^2 per day of glucose 5% and normal saline (1:1, per volume) was given to all patients starting 12 h before and continued until 48 h after the end of CDDP infusion. Two millimoles per kilogram KCl, 2 ml/kg Ca-gluconate (10%), 1 ml/kg Mg 20% and 0.4–0.8 ml/kg glycerophosphate (1 mmol P per ml) were added to the daily basal solution. Furthermore, mannitol 15% (40 ml/m^2) was administered as an i.v. bolus infusion 1 h before starting CDDP and if diuresis decreased significantly during treatment course (urine excreted $< 2/3$ of total fluid intake).

Blood and urine specimens

Blood samples were obtained from all patients before starting CDDP therapy, immediately after completion of CDDP administration, several times within the following 8 h and at 24 h after initiation of CDDP therapy. For the continuous infusion regimen, additional blood samples were drawn during CDDP administration. No exact time schedule could be maintained for reasons of patient care. Blood was collected in heparinised tubes and immediately centrifuged for 3 min at 14,000 rpm. Plasma was removed and 500 µl plasma was added to 50 µl 70% perchloric acid. The tubes were shaken and after centrifugation at 14,000 rpm for 5 min the supernatant was withdrawn. Plasma and deproteinated samples were frozen at –20 C until analysis of total and free Pt. We have evaluated the reliability of the deproteinating procedure using perchloric acid in an in vitro assay. Free Pt concentrations of CDDP-preincubated plasma samples (250 ng/ml, 500 ng/ml and 1000 ng/ml) were analysed after ultrafiltration using an Amicon ultrafiltration system (MW 10,000 cut-off, Amicon, Danvers, Mass.) and after acid precipitation as described above. The mean Pt concentrations measured using perchloric acid precipitation amounted to 90% of the values after ultrafiltration and variation coefficients were low ($< 6\%$). There was no concentration dependency using this procedure within the concentration range of the calibration curve (see below).

Urine specimens were collected as frequently as possible in order to detect peak urine CDDP concentrations. Small aliquots were frozen at –20 C for Pt analysis and the remaining urine was pooled as 24-h urine collections for calculation of creatinine clearances and determination of nephrotoxicity markers.

Pt analyses

Deproteinated plasma and urine were analysed for Pt using flameless atomic absorption spectroscopy at 2650 C and a wavelength of 265.9 nm using a GBC 904 AA spectrometer (Maassen, Ravensburg, Germany). If necessary, samples of urine and deproteinated plasma were diluted with water. The lower quantitation limit was 0.03 µg/ml and a standard curve from 0.03 µg/ml to 1 µg/ml was established with each assay using aqueous Pt standards. Recalibration was performed after analysis of ten urine or four plasma specimens. Intra- and inter-assay coefficients of variation were less than 5% for urine and diluted plasma and less than 10% for undiluted plasma.

Pharmacokinetic analysis

Pharmacokinetic parameters were determined for free Pt by fitting plasma concentrations and renal elimination of Pt to a two-compartment model using the Topfit computer model [26, 27]. The following parameters for free Pt were calculated: initial half-life ($t_{1/2}\alpha$), terminal half-life ($t_{1/2}\beta$), maximal concentration in plasma (C_{max}), AUC, volume of distribution in steady state (V_{SS}), total clearance (Cl_t), renal clearance (Cl_r) and the cumulative amount excreted in urine from time zero to infinity (A_e).

Nephrotoxicity

Concentrations of standard serum parameters (sodium, potassium, calcium, magnesium, phosphate, creatinine, blood urea nitrogen) were obtained from routine specimen withdrawn before beginning CDDP treatment and daily during treatment. Analyses were performed using a Beckman auto-analyser (Synchron CX5D, Beckman, München, Germany). Urinary nephrotoxicity markers were determined using 24-h urine specimen collected the day before CDDP infusion and from pooled urine samples collected during the pharmacokinetic study. Between CDDP courses (interval) and after completion of chemotherapy, collected (24-h) or spontaneous urine specimens (second morning urine) were used. Mean follow-up was 8.3 ± 5.9 months. Special care was taken by the parents and the staff to guarantee completeness of collection. The concentrations of urinary marker proteins for glomerular and tubular function [total urinary protein (t $Prot_U$), immunoglobulin G (IgG), transferrin (TRF), urinary albumin (Alb_U), α1-microglobulin (α1-M), retinol-binding protein (RBP) and *N*-acetyl-β-D-glucosaminidase (β-NAG)] were analysed using a Behring BNA nephelometer (840 nm, Behring, Marburg, Germany). Tamm-Horsfall protein (THP) was analysed using synelisa (synchron enzyme linked immuno sorbent assay, Elias, Freiburg, Germany). Proteinuria was analysed qualitatively by denaturing sodium dodecylsulfate-polyacrylamide gel electrophoresis (SDS-PAGE). The following concentrations were considered as pathological: Alb_U at least 20 mg/g $crea_U$, α1-M at least 15 mg/g $crea_U$ and RBP greater than 1 mg/l. Glomerular filtration rate (GFR) was estimated by the creatinine clearance [28] calculated from 24-h urine collections using the standard formula: $GFR\ [(ml/min)/1.73\ m^2] = (Crea_U \cdot Vol_U \cdot 1.73\ m^2)/(Crea_S \cdot time_C \cdot BSA)$ in which $Crea_U$ and $Crea_S$ are the concentrations of creatinine in urine and serum (mg/dl), Vol_U is the volume of urine excreted (ml), $time_C$ is the collection time (min) and BSA is the individual body surface area (m^2).

Statistics

Data are given as means (±SD). The Student's *t*-test for paired data was used to compare individual nephrotoxicity markers before and during CDDP administration as well as during the follow-up period. Relationships between infusion schedules and pharmacokinetic parameters as well as between pharmacokinetic parameters and nephrotoxicity markers were tested by calculating Pearson's correlation coefficient.

Results

Pharmacokinetics

The pharmacokinetic parameters obtained for free Pt are summarised in Table 2. Since different numbers of cycles per patient have been studied, statistical analysis was performed using all 25 CDDP cycles investigated as well as only one representative cycle per patient. For the latter, the cycles with the best correlation between the values measured and the calculated optimised parameters in the computer model were selected. There were only minor differences in the results between these two approaches (Table 2). After cessation of CDDP infusion, a bi-exponential decline of plasma free Pt concentrations was found in all treatment regimens with a mean initial half-life ($t_{1/2}\alpha$) of 21.6 ± 9.6 min and a

Table 2 Pharmacokinetic parameters of free (non-protein bound) platinum obtained by fitting results of plasma and urine analyses to a two compartment model. Statistical analysis was performed using one representative course (with the best correlation between the values measured and the calculated optimised parameters in the computer model) per patient ($n = 12$) and using all courses studied ($n = 25$). *Dose* cisplatin dose of the representative course, $t_{1/2}\alpha$ initial half-life, $t_{1/2}\beta$ terminal half-life, Cl_r renal clearance, Cl_t total clearance, A_e cumulative amount of platinum excreted in urine from time zero to infinity, V_{SS} volume of distribution, *AUC* area under the plasma concentration–time curve from time zero to infinity, C_{max} peak plasma platinum concentration

Patient	Infusion (h)	Dose (mg)	$t_{1/2}\alpha$ (h)	$t_{1/2}\beta$ (h)	$Cl^t \frac{[ml/min]}{m^2}$	$Cl^r \frac{[ml/min]}{m^2}$	A_e (mg/m^2)	A_e (% of dose)	V_{SS} (l/m^2)	AUC $\frac{h \cdot [\mu g/ml]}{[100mg/m^2]}$	C_{max} (μg/ml)
1	1×6	125	0.44	27.6	106	58.1	41.0	54.7	80.2	15.7	1.44
2	72	204	0.05	7.63	93.5	41.5	53.1	44.2	37.5	17.8	0.16
3	72	102	0.65	15.2	109	40.2	44.2	36.9	65.8	15.3	0.22
4	72	210	0.13	32.3	88.9	38.0	50.0	42.8	124	18.3	0.06
5	72	120	0.27	39.0	201	110	55.1	54.7	253	8.3	0.11
6	3×1	150	0.48	17.3	143	56.1	37.2	39.1	94.2	9.2	1.69
7	3×1	120	0.37	13.6	100	36.4	42.3	36.3	45.0	16.2	3.59
8	3×1	186	0.39	21.8	168	60.5	42.9	36.0	161	9.9	1.71
9	3×1	99	0.47	29.7	182	73.4	48.1	40.3	137	9.1	2.04
10	3×1	96	0.41	68.0	185	75.1	48.5	40.7	358	9.0	1.98
11	3×1	105	0.30	26.5	70.2	26.4	44.8	37.5	85.3	23.6	4.17
12	5×1	150	0.38	11.8	174	65.7	37.8	37.8	65.1	9.6	
Mean (±SEM), $n = 12$			0.36 ± 0.16	25.87 ± 16.21	135 ± 45.3	56.8 ± 22.8	45.4 ± 5.7	41.7 ± 6.6	126 ± 94.2	13.5 ± 4.97	1.52 ± 1.34
Mean (±SEM), $n = 25$			0.31 ± 0.17	26.72 ± 17.99	135 ± 58.4	54.0 ± 23.4	44.9 ± 6.9	40.9 ± 5.7	122 ± 77.8	15.5 ± 9.1	1.52 ± 1.44

terminal half-life ($t_{1/2}\beta$) of 25.9 ± 16.2 h ($n = 12$). Time courses of the free Pt concentration in plasma during the single 6-h infusion, the consecutive 1-h infusions and the 72-h continuous infusion are shown in Fig. 1A–C. Peak plasma concentrations of free Pt were observed at the end of the respective infusion times. The highest urinary Pt concentrations (Fig. 1D–F) and the maxima of urinary excretion rates (not depicted) were also found at that time. As expected, increased delivery rates of CDDP due to higher doses or more rapid i.v. infusion were followed by higher peak Pt concentrations in plasma and urine as well as by higher urinary excretion rates (Fig. 2A–C). The correlation coefficients were 0.79, 0.73 and 0.69, respectively ($P < 0.001$). Even though short collection periods for urinary sampling were chosen, the maximal urinary Pt levels were most likely underestimated in the treatment groups using a 1-h infusion schedule, but urinary catheterisation of the patients to obtain more detailed results was considered inappropriate. The A_e revealed only small inter-individual variations. Approximately 42% of the injected doses was eliminated in the urine. But A_e also was influenced by the CDDP delivery rate (correlation coefficient = –0.54, $P < 0.01$, Fig. 2E). In contrast to this, a greater variability was seen with the mean normalised Pt clearances, which were not related to the infusion schedules. Total clearance exceeded the renal clearance about 2.4-fold, indicating that additional pathways of Pt elimination as reactions with endogenous macromolecules must be taken into account. Furthermore, renal Pt clearance values were lower than the creatinine clearances obtained on the first day of CDDP treatment of the same cycle (ratio 0.6 ± 0.2). Mean AUC of free cisplatin averaged 13.5 ± 5.0 μg/ml·h per 100 mg/m^2 with considerable variations within the same dosing schedule. There was a slight tendency towards lower AUC levels when CDDP was given as repetitive 1-h bolus than as a continuous 72-h infusion, but there was no significant relationship between the CDDP delivery and the AUC (Fig. 2D, correlation coefficient = –0.19, $P > 0.05$). Since the amount of free CDDP is considered to be responsible for both tumour response and organotoxic side effects, the substantial inter-individual differences in AUC observed using equal doses need further evaluation in order to optimise CDDP regimens.

Nephrotoxicity

All children had normal nephrotoxicity markers before the first CDDP administration, except for one patient with unknown basal creatinine clearance (not performed). During CDDP treatment, serum creatinine, electrolyte and calcium concentrations remained within normal ranges whereas hypomagnesaemia and hypophosphataemia were observed in several patients. Due to a nonuniform substitution therapy with magnesium and phosphate, both being given for prophylactic as well as for therapeutic purposes, no quantitative evaluation of

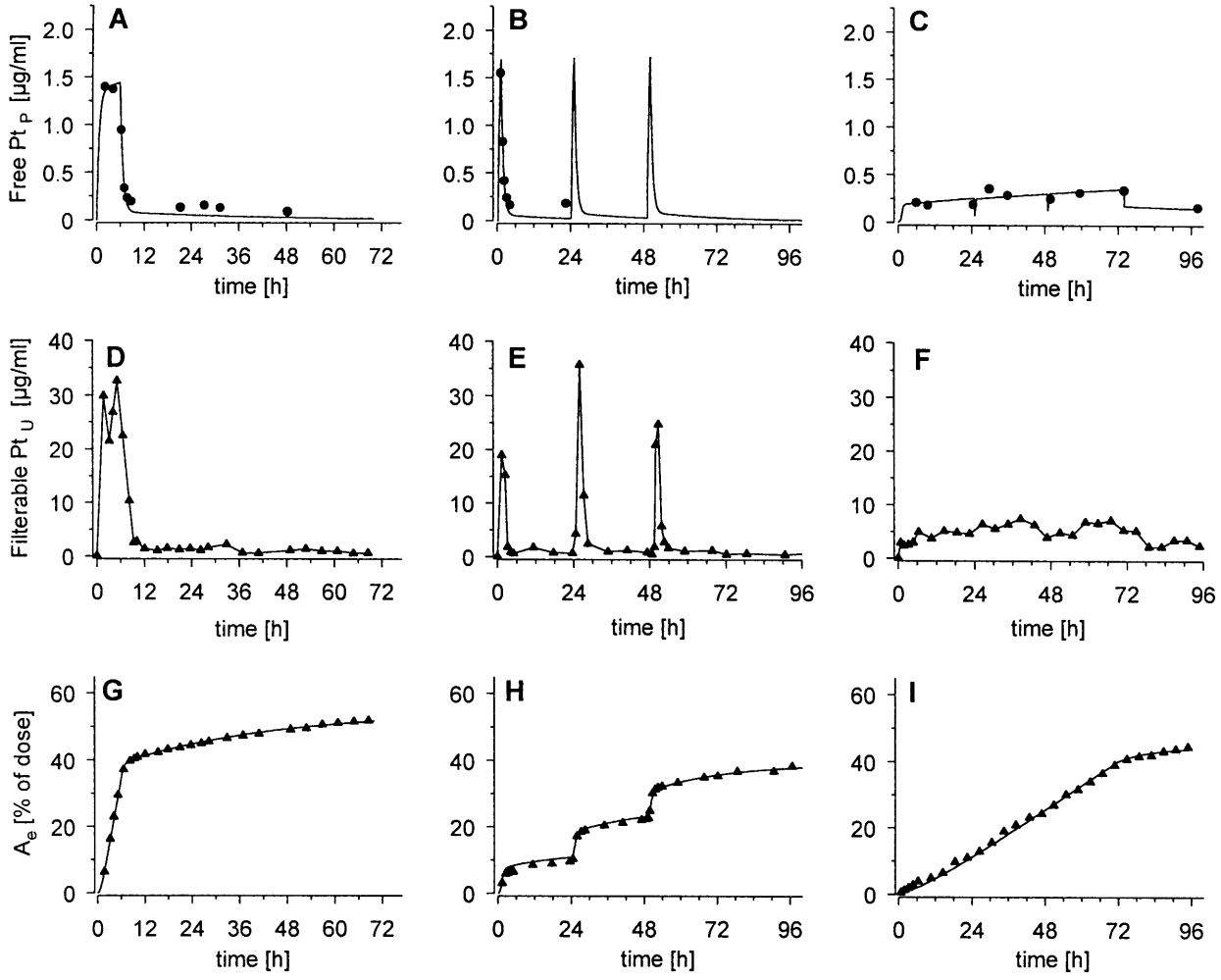

Fig. 1 Time courses of free platinum concentrations in plasma (Pt_P) and urine (Pt_U) and of cumulative urinary platinum excretion (A_e) under the different infusion regimens. Single 6-h infusion of 75 mg/m^2 (*left*, A, D, G; patient 1, cycle 1), three consecutive 1-h infusions (3×40 mg/m^2) within 72 h (*middle*, B, E, H; patient 6, cycle 1) and 72-h continuous infusion of 120 mg/m^2 (*right*, C, F, I; patient 2, cycle 2)

CDDP-induced urinary loss of magnesium or phosphate was performed.

In contrast to serum parameters, urinary nephrotoxicity markers provided clear evidence for glomerular and tubular damage following CDDP administration. The quantitative analysis of urinary marker proteins is shown in Table 3. Maximal average daily protein loss under CDDP was 440 mg, which was equivalent to a 2.6-fold increase of daily urinary protein excretion compared with the pre-infusion levels ($P < 0.05$). The glomerular markers IgG and TRF were always below the detection limit before starting CDDP, whereas a moderate and reversible increase of both urinary IgG and TRF concentrations was seen during CDDP therapy. The creatinine clearance (GFR) showed a significant CDDP-associated decrease during the treatment courses (Table 3, $P < 0.05$). The mean nadir under CDDP therapy corresponded to 70% of baseline values. As for tubular marker proteins, a significant increase in α1-M and albumin excretion was detected in all courses investigated (Table 3, $P < 0.05$). The activity of the tubular marker enzyme β-NAG and the excretion of THP also revealed marked and significant changes following CDDP administration (Table 3) whereas RBP increased above the detection limit in only five courses (data not shown). These findings provide evidence for CDDP-induced damage at different levels of the tubular system. The pathological values gradually returned to normal levels until the subsequent cisplatin course. After cessation of CDDP, however, a subclinical proteinuria persisted in a considerable number of patients until a mean follow-up of 8 months, as indicated by pathological findings using denaturing SDS-PAGE in 50% of patients.

The quantitative relationship between the pharmacokinetics of unbound CDDP and its associated nephrotoxicity was examined by calculating the correlation coefficients between free CDDP pharmacokinetic parameters and urinary markers for renal function. As shown in Table 4, pharmacokinetic results provided only limited information about the severity of cisplatin-induced nephrotoxic side effects. Predictive parameters were the peak concentrations of free CDDP in plasma

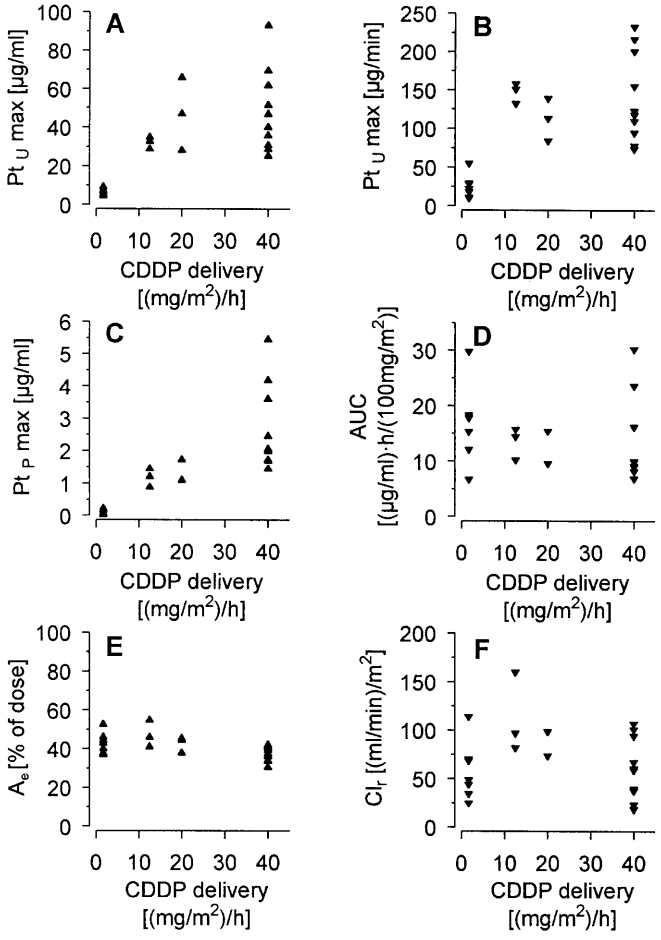

Fig. 2 Relationship between normalised cisplatin (CDDP) delivery and pharmacokinetic parameters. Positive correlation between CDDP delivery given as $(mg/m^2)/h$ and peak free platinum concentrations in urine (A) and plasma (C) as well as peak urinary excretion rate of platinum (B); $P < 0.05$. Negative correlation between CDDP delivery and cumulative elimination of platinum in urine A_e ($P < 0.05$, E). No significant correlation between CDDP delivery and area under the concentration–time curve (AUC, D) or renal platinum clearance (Cl_r, F)

and urine and the cumulative excreted amount of Pt in urine (Fig. 3). The extent of reduction in GFR and of increases in urinary excretion of albumin and β-NAG were positively correlated with peak Pt concentrations in plasma and urine, whereas a negative correlation of the decrease in GFR and the increase of urinary albumin excretion was found with the cumulative urinary elimination of Pt (Fig. 3, Table 4, $P < 0.05$).

Discussion

The paucity of literature dealing with the pharmacokinetics of CDDP in childhood stands in stark contrast to the frequent use of this drug in paediatric cancer patients and the severity of its toxicity. Since there is a variety of difficulties to overcome in conducting pharmacokinetic investigations in children, available data have been derived mostly from the results of studies in adults, which were extrapolated even to very young patients. Therefore, the goal of the present study was to provide a broad spectrum of pharmacokinetic data of CDDP in paediatric patients, to document the incidence and severity of CDDP associated nephrotoxicity and to evaluate the relationship between pharmacokinetic parameters and CDDP-induced renal impairment.

The pharmacokinetic parameters for free CDDP were obtained from different CDDP infusion schedules, with 10 of 12 patients receiving doses of 120 mg/m^2, one patient receiving 100 mg/m^2 and another patient 75 mg/m^2 CDDP per course. The rate of drug delivery had a significant effect on the peak free Pt concentrations in plasma, leading to higher maximal concentrations with more rapid infusion rates. As a consequence, the maximal urine concentrations of Pt and the peak urinary excretion rate of Pt were also elevated (Fig. 2). Furthermore, the total administered dose per cycle was related to peak free plasma Pt ($P < 0.05$, data not shown).

Since peak free plasma and urinary Pt concentrations were significantly related to the decrease in GFR and to the increase in urinary albumin and β-NAG excretion during CDDP treatment (Fig. 3), our results provide evidence that CDDP dose rate influences the extent of associated glomerular and tubular toxicity. Recently published data by Skinner and co-workers [29] also have shown that the severity of CDDP nephrotoxicity in children is related to the cisplatin dose rate. The relationship between plasma Pt concentrations and CDDP-induced nephrotoxicity has been described by pharmacokinetic studies in adults [12, 13, 14]. Given that the rate of CDDP administration appears to be a determinant of nephrotoxicity in children in our study, our results support the view already derived from adults [15, 16, 17, 30], that continuous infusion schedules should be favoured whenever CDDP is administered to cancer patients due to the lower associated toxicity and preserved antitumour efficacy.

The dependence of the AUC on the CDDP infusion rate is debated in the literature. Higher AUC levels were described to be associated with continuous CDDP infusions [19, 31], whereas Vermorken and associates [20] reported similar AUCs with 8-min, 3-h and 24-h infusions. We found a higher AUC with the continuous infusion than the intermittent 1-h infusions at the same dose range, but these differences were not statistically significant. The dose-normalised AUC levels for free CDDP in our study revealed a high inter-individual variability (up to 2.8-fold) even if identical doses and infusion schedules were compared (Table 2). The phenomenon of high inter-individual AUC variability in children has already been described by Peng et al. [23] in a pharmacokinetic study including 21 patients receiving a 24-h infusion of CDDP, indicating that surface area-based dosing of CDDP is not satisfactory for optimal use of this drug in children. Murakami et al. [22] described age-dependent ototoxicity following CDDP

Table 3 Urinary markers for cisplatin (CDDP)-induced nephrotoxicity. *α1-M* α1-microglobulin, *Alb_U* urinary albumin, *t $Prot_U$* urinary total protein, *β-NAG* N-acetyl-β-D-glucosaminidase, *THP* Tamm-Horsfall-protein, *IgG* immunoglobulin G, *TRF* transferrin, *GFR* glomerular filtration rate estimated using creatinine clearance (calculated from 24-h urine collections). Mean follow-up was 8.3 ± 5.9 months

	GFR $\frac{[ml/min]}{m^2}$	α1-M (mg/g crea)	Alb_U (mg/g crea)	t $Prot_U$		β-NAG (U/g crea)	THP (mg/24 h)	IgG (mg/l)	TRF (mg/l)
				mg/g crea	mg/24 h				
Before first CDDP course ($n = 12$)	116 ± 25.7	13.1 ± 8.0	17.8 ± 6.5	204 ± 112	85.5 ± 38.1	5.0 ± 2.5	13.3 ± 13.7	< 3.8	< 2.4
At start of course ($n = 25$)	111 ± 29.5	15.4 ± 12.8	22.5 ± 14.5	231 ± 107	168 ± 106	5.0 ± 2.7	12.8 ± 12.0	< 3.8	< 2.4
Max or min during course ($n = 25$)	77.9 ± 20.7***	83.1 ± 64.2***	136 ± 142*	737 ± 705***	440 ± 247*	71.8 ± 95.9**	1.60 ± 1.91***	7.77 ± 3.97*	4.70 ± 2.29*
Follow up ($n = 12$)	104 ± 20.6	10.0 ± 5.1	15.1 ± 8.1	117 ± 80.2	101 ± 77.8	4.0 ± 2.4	26.5 ± 19.8	< 3.8	< 2.4

*$P < 0.05$
**$P < 0.01$
***$P < 0.001$, during course vs start course

treatment and concluded from their pharmacokinetic data that the higher toxicity in younger children was associated with both a lower clearance rate and a higher distribution volume than in older children. In our study, however, a relation between AUC levels and nephrotoxicity markers could not be established (Table 4). Nevertheless, the high variability of CDDP excretion parameters in children needs further evaluation in order to optimise dose regimen.

The total free Pt clearance exceeded renal clearance about 2.4-fold in our patients, indicating that elimination pathways other than renal excretion contribute to inter-individual variability of available Pt. In contrast to the results of Dominici et al. [25] our study results show no tendency towards increasing AUC levels with consecutive CDDP courses in the same patient. On the contrary, a decrease in AUC and a higher concomitant Pt clearance was found when analysing consecutive cycles (data not shown), but in view of the small number of patients these data should be interpreted with caution.

In all courses, signs of CDDP-associated nephrotoxicity were observed. The urinary excretion of α1-M and albumin (related to urinary creatinine concentration) turned out to be the most sensitive markers for the detection of acute renal tubular damage, revealing pathological findings in 100% of CDDP treatments. The plasma concentrations of creatinine and blood urea nitrogen which are the standard parameters for monitoring renal function in clinical practise, however, remained unchanged throughout all CDDP courses. Thus, plasma creatinine concentrations were not reliable indicators of renal toxicity, which had already been demonstrated by other groups [6, 7, 32]. However, the creatinine clearance significantly decreased following CDDP infusion. Although methodological objections have been raised in the past against using creatinine clearance due to unreliability and tendency to overestimate GFR [7, 32], our

Table 4 Correlation coefficients between pharmacokinetic parameters of free cisplatin (CDDP) and urinary nephrotoxicity markers. *n* varied between 20 and 25 courses. *$C_{max\ P}$* peak plasma concentration of free platinum, *$C_{max\ U}$* peak urine concentration of filterable Pt, *A_e* cumulative amount of Pt excreted in the urine from time zero to infinity, *Cl_r* renal Pt clearance, *Cl_t* total Pt clearance, *AUC* area under the plasma concentration–time curve from time zero to infinity, *α1-M max* maximal concentration of α-1-microglobulin in the urine, *β-NAG max* maximal concentration of N-acetyl-β-D-glucosaminidase in the urine, *THP min* minimal urinary concentration of Tamm-Horsfall protein, *GFR min* minimal glomerular filtration rate (estimated using creatinin clearance), *r* correlation coefficient (Pearson's correlation), *NS* not significant

		α1-M max (mg/g crea)	Alb_U max (mg/g crea)	t $Prot_U$ max (mg/g crea)	β-NAG max (U/g crea)	THP min (mg/24 h)	GFR min $\frac{[ml/min]}{1.73\,m^2}$
$C_{max\ P}$	r	0.397	0.504	0.05	0.502	–0.277	–0.552
µg/ml	P	NS	0.017	NS	0.017	NS	0.012
C_{maxU}	r	0.127	0.412	0.043	0.487	–0.214	–0.462
µg/ml	*P*	NS	0.046	NS	0.014	NS	0.040
A_e	r	–0.308	–0.507	–0.229	–0.299	–0.321	0.585
% of dose	*P*	NS	0.011	NS	NS	NS	0.007
Cl_r	r	–0.402	–0.241	–0.1	–0.296	–0.065	0.224
(ml/min)/m^2	*P*	NS	NS	NS	NS	NS	NS
Cl_t	r	–0.328	-0.08	–0.037	–0.224	0.075	–0.02
(ml/min)/m^2	*P*	NS	NS	NS	NS	NS	NS
AUC	r	0.251	0.013	–0.076	0.127	0.220	0.063
h·(µg/ml)	*P*	NS	NS	NS	NS	NS	NS

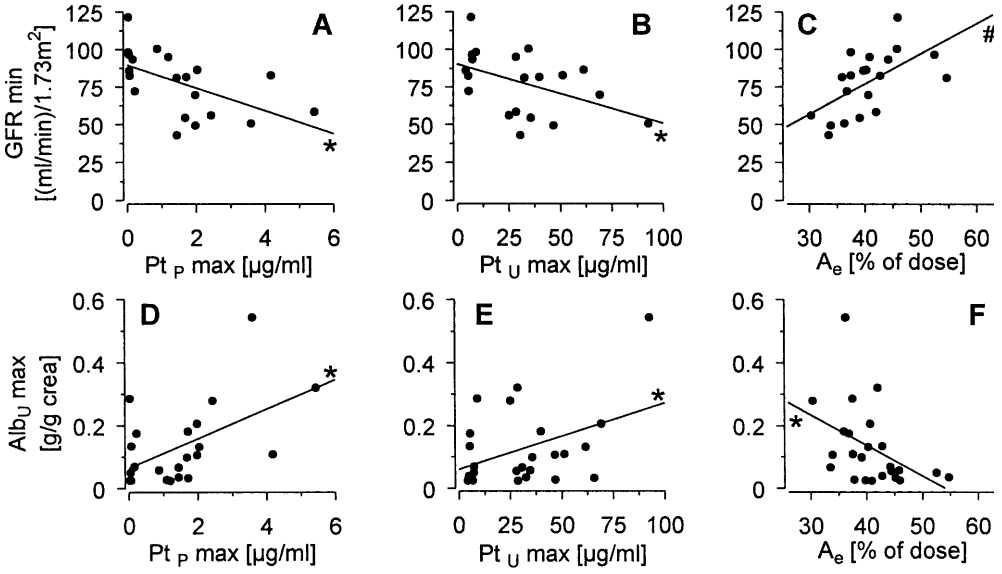

Fig. 3 Relationship between pharmacokinetic parameters and cisplatin (CDDP)-induced nephrotoxicity. Dependence of minimal glomerular filtration rate (GFR) value (*upper panel*) and peak urinary concentrations of albumin (Alb_{max}, *lower panel*) on pharmacokinetic parameters during CDDP treatment. $Pt_{P\ max}$ peak free Pt concentrations in plasma, $Pt_{U\ max}$ peak free Pt concentrations in urine, A_e cumulative urinary Pt elimination from time zero to infinity. Correlation coefficients were: –0.55 (A), –0.46 (B), 0.59 (C), 0.5 (D), 0.65 (E), –0.51 (F); Pearson's correlation, $n = 20$–24 courses, $^{*}P < 0.05$, $^{\#}P < 0.01$

results are in good agreement with those obtained using inulin clearance [33] or the ^{51}Cr-ethylene diamine tetraacetic acid (EDTA) clearance technique [5]. Renal dysfunction was only partially reversible after completion of the chemotherapeutic treatment, with five of nine patients evaluated revealing a subclinical microproteinuria 12 months after their last CDDP infusion. Furthermore, GFR remained slightly decreased during follow-up and a significant negative correlation was found between maximum urinary Pt concentrations during CDDP infusion and GFR during follow-up (Fig. 4, $P < 0.05$). In the literature, only few long-term follow-up studies have been performed to evaluate renal function after therapy with cisplatin. Divergent results have been obtained by different groups, varying from minor functional abnormalities [34, 35, 36] to mild renal dysfunction [5, 33, 37] to progressive deterioration of renal function [38, 39]. Since the vast majority of treatment schedules in paediatric oncology consists of combination chemotherapy often including other nephrotoxic drugs, such as ifosfamide or methotrexate, it is difficult to estimate the contribution of the individual drugs in the development of renal dysfunction and no general conclusions can be drawn from current paediatric studies. In our study, ifosfamide and methotrexate were also given to patients with osteosarcoma or medulloblastoma. In the latter group there was an interval of 12 weeks between ifosfamide and cisplatin bolus administration and the evaluation of the nephrotoxicity markers revealed no indication of relevant additional ifosfamide-induced nephrotoxicity in these patients. Osteosarcoma patients received ifosfamide concurrently with the long-term cisplatin infusion and hypophosphataemia probably due to concurrent ifosfamide was observed in a few patients. However, the excretion of urinary marker proteins showed significantly lower renal dysfunction in the continuous infusion group than the bolus infusion and assuming that ifosfamide contributes to nephrotoxicity the toxic effects of cisplatin might be overestimated in these cases.

The analysis of the quantitative relationship between pharmacokinetics of free CDDP and Pt-induced nephrotoxicity identified only the peak concentrations of free Pt in plasma and urine as well as the cumulative Pt amount excreted in urine to be predictive parameters for Pt-induced renal dysfunction. The C_{max} of free Pt already has been shown to correlate with the minimum creatinine clearance in a pharmacokinetic study in adults [11]. Furthermore, total plasma Pt concentrations were also related to CDDP-associated nephrotoxicity [12, 13]. Consistent with these results, our data provide further evidence that with respect to nephrotoxicity continuous cisplatin infusions may be superior to intermittent bolus

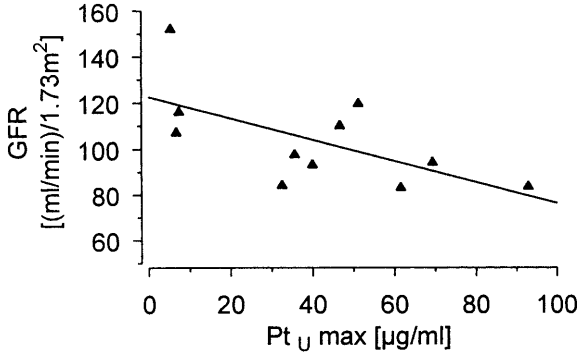

Fig. 4 Correlation between maximum free Pt concentrations in urine (Pt_{U} max) and glomerular filtration rate (GFR) during follow-up (8.3 ± 5.9 months). Correlation coefficient = –0.63, $P < 0.05$, $n = 11$ patients

administration because of lower Pt levels in plasma and urine.

In conclusion, no dramatic differences arose when comparing the pharmacokinetic parameters obtained from children in our study with the sparse data in the literature or those derived from adults. Subclinical CDDP-induced nephrotoxicity was found during all courses investigated with only partial reversibility after cessation of CDDP treatment. Lower CDDP delivery rates were less nephrotoxic and the excretion of urinary α1-M and albumin were the most sensitive markers for Pt-induced renal dysfunction.

Acknowledgements We thank Dr. Eberhard Schleyer for his advice about analysing pharmacokinetic parameters using the Topfit computer model. This work was supported in part by a grant from the Deutsche Forschungsgemeinschaft (SFB 330).

References

1. Green AA, Hayes FA, Pratt CB, Evans WE, Howarth CB, Senzer N (1980) Phase II evaluation of cisplatin in children with neuroblastoma and other malignant solid tumors. In: Prestayko AW, Crooke ST, Carter SK (eds) Current status and new developments with cisplatin. Academic Press, New York, pp 477–484
2. Pinkerton CR, Pritchard J, Spitz L (1986) High complete response rate in children with advanced germ cell tumors using cisplatin-containing combination chemotherapy. J Clin Oncol 4:194–199
3. Fillastre JP, Raguenz-Viotte G (1989) Cisplatin nephrotoxicity. Toxicol Lett 46:724–725
4. Krakoff IH (1979) Nephrotoxicity of cis-dichlorodiammine platinum (II). Cancer Treat Rep 63:1523–1525
5. Brock PR, Koliouskas DE, Barratt TM, Yeomans E, Pritchard J (1991) Partial reversibility of cisplatin nephrotoxicity in children. J Pediatr 118:531–534
6. Daugaard G, Abilgaard U (1989) Cisplatin nephrotoxicity. Cancer Chemother Pharmacol 25:1–9
7. Skinner R, Pearson ADJ, Coulthard MG, Skillen AW, Hodson AW, Goldfinch ME, Gibb I, Craft AW (1991) Assessment of chemotherapy-associated nephrotoxicity in children with cancer. Cancer Chemother Pharmacol 28:81–92
8. Ozols RF, Cordon BJ, Jacobs J, Wesley M, Ostchega Y, Young RC (1984) High-dose cisplatin in hypertonic saline. Ann Intern Med 100:19–24
9. Howell SB, Pfeifle CE, Wung WE, Olshen RA (1983) Intraperitoneal cis-diaminedichloroplatinum with systemic thiosulfate protection. Cancer Res 43:1426–1431
10. Pfeifle CE, Howell SB, Felthouse RD, Woliver TBS, Andrews PA, Markman M, Murphy MP (1985) High-dose cisplatin with sodium thiosulfate protection. J Clin Oncol 3:237–244
11. Nagai N, Kinoshita M, Ogata H, Tsujino D, Wada Y, Someya K, Ohno T, Masuhara K, Tanaka Y, Kato K, Nagai H, Yokoyama A, Kurita Y (1996) Relationship between pharmacokinetics of unchanged cisplatin and nephrotoxicity after intravenous infusions of cisplatin to cancer patients. Cancer Chemother Pharmacol 39:131–137
12. Campbell AB, Kalman SM, Jacobs C (1983) Plasma platinum levels: Relationship to cisplatin dose and nephrotoxicity. Cancer Treat Rep 67:169–172
13. Kelsen DP, Alcock N, Young CW (1985) Cisplatin nephrotoxicity correlation with plasma platinum concentrations. Am J Clin Oncol 8:77–80
14. Reece PA, Stafford I, Russell J, Khan M, Gill PG (1987) Creatinin clearance as a predictor of ultrafilterable platinum disposition in cancer patients treated with cisplatin: relationship between peak ultrafilterable platinum plasma levels and nephrotoxicity. J Clin Oncol 5:304–309
15. Jacobs C, Bertino JR, Goffinet DR, Fee WE, Goode RL (1978) 24-hour infusion of cis-platinum in head and neck cancers. Cancer 42:2135–2140
16. Posner MR, Ferrari L, Belliveau JF, Cummings FJ, Wiemann MC, O'Rourke A, Weitberg AB, Calabresi P (1987) A phase I trial of continuous infusion cisplatin. Cancer 59:15–18
17. Salem P, Khalyl M, Jabboury K, Hashimi L (1984) cis-Diaminedichloroplatinum (II) by 5-day continuous infusion. A new dose schedule with minimal toxicity. Cancer 53:837–840
18. Bonetti A, Franceschi T, Apostoli P, Messori A, Speretto L, Cetto GL, Molino A, Leone R (1995) Cisplatin pharmacokinetics using a five-day schedule during repeated courses of chemotherapy in germ cell tumors. Ther Drug Monit 17:25–32
19. Forastiere AA, Belliveau JF, Goren MP, Vogel WC, Posner MR, O'Leary GP (1988) Pharmacokinetic and toxicity evaluation of five-day continuous infusion versus intermittent bolus cis-diaminedichloroplatinum (II) in head and neck cancer patients. Cancer Res 48:3869–3874
20. Vermorken JB, van der Vijgh WJF, Klein I, Gall HE, van Groeningen CJ, Hart GAM, Pinedo HM (1986) Pharmacokinetics of free and total platinum species after rapid and prolonged infusions of cisplatin. Clin Pharmacol Ther 39:136–144
21. Bues-Charbit M, Gentet J-C, Bernard J-L, Breant V, Cano J-P, Raybaud C (1987) Continuous infusion of high-dose cisplatin in children: pharmacokinetics of free and total platinum. Eur J Cancer Clin Oncol 23:1649–1652
22. Murakami T, Inoue S, Sasaki K, Fujimoto T (1990) Studies on age-dependent plasma platinum pk and ototoxicity of cisplatin. Sel Cancer Ther 6:145–151
23. Peng B, English MW, Boddy AV, Price L, Wyllie R, Pearson ADJ, Tilby MJ, Newell DR (1997) Cisplatin pharmacokinetics in children with cancer. Eur J Cancer 33:1823–1828
24. Crom WR, Evans WE, Pratt CB, Senzer N, Denison M, Green AA, Hayes FA, Yee GC (1981) Cisplatin disposition in children and adolescents with cancer. Cancer Chemother Pharmacol 6:95–99
25. Dominici C, Petrucci F, Caroli S, Alimonti A, Clerico A, Castello MA (1989) A pharmacokinetic study of high-dose continuous infusion cisplatin in children with solid tumors. J Clin Oncol 7:100–107
26. Heinzel G, Hammer R, Wolf M, Koss FW, Bozler G (1977) Model building in pharmacokinetics. Part III: Simplified rules for the deduction of analytical solutions for linear compartment models. Drug Res 27:904–911
27. Transwell P, Koup J (1993) TopFit: a PC-based pharmacokinetic/pharmacodynamic data analysis program. Int J Clin Pharmacol Ther Toxicol 10:514–520
28. Barrat TM (1974) Assessment of renal function in children. In: Apley J (ed) Modern trends in pediatrics, vol. 4. Butterworth, London, pp 181–215
29. Skinner R, Pearson ADJ, English MW, Price L, Wyllie RA, Coulthard MG, Craft AW (1998) Cisplatin dose rate as a risk factor for nephrotoxicity in children. Br J Cancer 77:1677–1682
30. Lokich JJ (1980) Phase I study of cis-diaminedichloroplatinum (II) administered as a constant 5-day infusion. Cancer Treat Rep 64:905–908
31. Belliveau JF, Posner MR, Ferrari L, Crabtree GW, Cummings FJ, Wiemann M, O'Leary GP, Griffin H, Phaneuf MA, O'Rourke A, Calabresi P (1986) Cisplatin administered as a continuous 5-day infusion: plasma platinum levels and urine platinum excretion. Cancer Treat Rep 70:1215–1217
32. Womer RB, Pritchard J, Barratt TM (1985) Renal toxicity of cisplatin in children. J Pediatr 106:659–663
33. Brillet G, Deray G, Jacquiraud C, Mignot L, Bunker D, Meillet D, Raymond F, Jacobs C (1994) Long-term renal effect of cisplatin in man. Am J Nephrol 14:81–84
34. Arndt C, Morgenstern B, Hawkins D, Wilson D, Liedtke R, Miser J (1999) Renal function following combination chemotherapy with ifosfamide and cisplatin in patients with osteogenic sarcoma. Med Pediatr Oncol 32:93–96

35. Fjeldborg P, Sørensen J, Helkjær PE (1986) The long-term effect of cisplatin on renal function. Cancer 58:2214–2217
36. Hamilton CR, Bliss JM, Horwich A (1989) The late effects of cis-platinum on renal function. Eur J Cancer Clin Oncol 25:185–189
37. Koch Nogueira PC, Hadj-Aissa A, Schell M, Dubourg L, Brunat-Mentigny M, Cochat P (1998) Long-term nephrotoxicity of cisplatin, ifosfamide, and methotrexate in osteosarcoma. Pediatr Nephrol 12:572–575
38. Brillet G, Deray G, Lucsko M, Faucher C, Aubert P, Rottembourg J, Jacobs C (1993) Insuffisance rénale chronique terminale définitive après traitement par le cisplatine. Néphrologie 14:227–229
39. Guinee DG, Van Zee B, Houghton DC (1993) Clinically silent progressive renal tubulointerstitial disease during cisplatin chemotherapy. Cancer 71:4050–4054