Cancer Chemother Pharmacol (1988) 22: 11–16

Cancer Chemotherapy and Pharmacology

The comparative tissues distribution of platinum and ^{14}C in mice receiving ^{14}C-labelled carboplatin*

Sarah E. Dible, Zahid H. Siddik, and Kenneth R. Harrap

Drug Development Section, The Institute of Cancer Research, Sutton, Surrey SM2 5PX, England

Summary. Since the reactivity of carboplatin depends on the rate of removal of the 1,1-cyclobutanedicarboxylate ligand, the time course of this dissociation has been determined in various tissue and body fluids using ^{14}C-labelled carboplatin (*cis*-diammine[1,1-cyclobutane-1-^{14}C-dicarboxylate]platinum II). Mice received ^{14}C-carboplatin (80 mg/kg; 1.1 mCi/kg, i.v.), and tissue was removed at times ranging from 5 min to 5 days posttreatment. Following solubilization, tissue aliquots were analyzed for platinum and ^{14}C contents. Carboplatin remained intact for up to 2 h posttreatment, since the ratio of ^{14}C:Pt in tissues (nmol/g) was unity. Thereafter, the ^{14}C ligand was released from the molecule and preferentially removed from tissues, indicated by decreasing ^{14}C:Pt ratios. The elimination half-lifes for Pt varied between tissues (40–156 h). In contrast, the corresponding half-lifes for the ^{14}C species were similar in most types of tissue (18–35 h), although those in the liver and spleen were excetpional (210 and 90 h, respectively). At 5 days a maximum of 4%–24% of the total Pt in tissue might exist as intact drug. Thus, the metabolic handling of carboplatin varies according to the tissue, since the elimination of the ^{14}C cyclobutane dicarboxylate species from most tissue was similar and Pt elimination was slower and tissue-dependent.

Introduction

Cisplatin (*cis*-dichlorodiammine platinum II) is a well-established agent in cancer chemotherapy, although its side effects, notably dose-limiting nephrotoxicity, have hindered its full clinical exploitation [9, 11, 17]. Carboplatin was developed as a more stable, non-nephrotoxic analogue of cisplatin [5]. Phase I evaluation revealed myelosuppression to be dose-limiting and confirmed an absence of nephrotoxicity at the recommended phase II dose [1]. Subsequent clinical studies in other centers have confirmed these findings [4]. Phase II studies have revealed activity in ovarian [23], small cell lung [22], and testicular [16] tumors, and the drug was registered in the United Kingdom for the two former indications in 1986. The differential nephrotoxicities of cisplatin and carboplatin have been attributed to differences in the mechanisms of their renal handling [20]. The chemical reactivity of the complex may also be an important determinant of nephrotoxicity [3]. However, both cisplatin and carboplatin are inert as intact molecules, and their activation must occur before any biological activity is manifested. The activation of cisplatin is chemically mediated and requires aquation of the molecule [10], the first aquation step probably being rate-limiting in the platination of macromolecules [3, 10]. Carboplatin is thought to be activated in a similar manner, although the presence of the bidentate cyclobutane dicarboxylate ligand, in place of the two chloride ligands of cisplatin, renders the former compound 10- to 20-fold less reactive [8, 20]. This, however, does not discount the possibility of enzyme-mediated activation of carboplatin, as proposed by Cleare [2]. The rate of activation in a tissue may be related to its chemical and biochemical composition, since we have previously demonstrated that the binding of both carboplatin and cisplatin in the liver and kidney in vitro is significantly greater than that in the plasma [20].

In the present investigation, we have examined the abilities of tissue to activate carboplatin in vivo. This was achieved through synthesis of ^{14}C-carboplatin, where the label is present in the cyclobutane dicarboxylate ligand (Fig. 1), and by following separately the pharmacokinetics of Pt and ^{14}C species.

Materials and methods

Chemicals

Carboplatin (*cis*-diamminecyclobutanedicarboxylate platinum II, JM8, Paraplatin) a gift from the Johnson Matthey Research Centre (Reading, Berks, UK) was dissolved in 5% dextrose. ^{14}C-cyclobutane dicarboxylic acid was obtained from Amersham UK (Amersham, Bucks, UK). Hyamine hydroxide (methylbenzethonium hydroxide in 1 *M* methanol) was obtained from Sigma Chemicals (UK).

Methods

^{14}C-carboplatin synthesis. 1,1-cyclobutane-^{14}C-dicarboxylic acid was synthesized by Amersham (UK) and had a specific activity of 68 μCi/mg. ^{14}C-carboplatin was synthesized

* The research reported was supported by grants to The Institute of Cancer Research: Royal Cancer Hospital from the Cancer Research Campaign and the Medical Research Council. SED thanks the Cancer Research Campaign and the Johnson Matthey company for their support. This work has been published, in part, in "Biochemical Mechanisms of Anticancer Drugs" (McBrien & Slater, eds) 1986

Offprint requests to: S. E. Dible

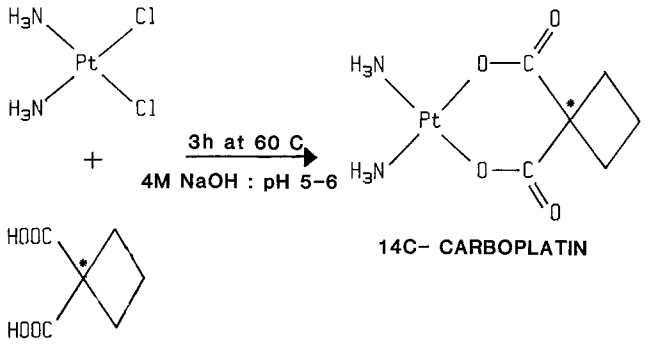

Fig. 1. Synthesis of *cis*-diammine-1,1-cyclobutane-1-^{14}C-dicarboxylate platinum II. * indicates the position of the ^{14}C atom

as described by the Research Corporation [7], as shown in Fig. 1. Briefly, silver nitrate (3.2 mmol) was dissolved in 1 ml distilled water. Platinum diiodiammine (1.66 mmol) was added with rapid stirring and maintained at 40° C for 1 h in a dark environment. The silver iodide precipitate was filtered off, and activated charcoal was added to the filtrate with 4 × 0.5 ml rinses of distilled water. ^{14}C-cyclobutane dicarboxylic acid (2.11 mmol) was dissolved in 0.75 ml 4 *M* sodium hydroxide, the final solution having a pH of 5–6. The diaquo Pt filtrate was added to ^{14}C-cyclobutane dicarboxylic acid with 0.5 ml water and the reaction maintained at 60° C for 3 h with formation of a white precipitate. The temperature was closely monitored to prevent decomposition of the product (> 60° C). The white precipitate was isolated by filtration, rinsed with 1 ml water and ethanol, and then reheated to 60° C in a flask with 1 ml distilled water. The solution was refiltered into a flask for freeze-drying over 24 h. The crude yield (52% yield) of ^{14}C-carboplatin was 0.87 mmol.

Analysis of ^{14}C-carboplatin purity. The chemical purity of the compound (> 96%) was assessed using flameless atomic absorption spectrophotometry. Its radiochemical purity was assessed by spotting 0.2 nmol ^{14}C-carboplatin (in water) onto silica gel-treated (60 F_{254}) thin-layer chromatography plates (Merck, Germany) and chromatographing for 30 min in acetone:water (4:1). After drying, the plates were scanned at 254 nm using a computerized Bertrol system, and 98.56% radioactivity (^{14}C-carboplatin) was located in a single peak. The specific activity of the product was assessed by liquid scintillation counting, using 8 ml cocktail T scintillant (BDH, UK) and ^{14}C-hexadecane as the external standard (Amersham, UK; specific activity 0.39 μCi/ml). The counting efficiency was > 96%. The sepcific activity of ^{14}C-carboplatin was 24.07 μCi/mg (4.66 μCi/mmol).

Tissue distribution studies. Female Balb/C− mice (18–20 g) received 20–25 μCi of ^{14}C-carboplatin (80 mg/kg; 1.1 mCi/kg; 10 ml/kg) or ^{14}C-cyclobutane dicarboxylic acid (31 mg/kg; 1.1 mCi/kg; 10 ml/kg). The dose solutions comprised radiolabelled drug diluted to the required dose with "cold" unlabelled drug in order to obtain the specific activities quoted.

Mice were anesthetized with diethylether and exsanguinated at 5 min, 1, 4, and 8 h, 1, 2, and 5 days post treatment. Tissue (liver, kidney, muscle, ileum, heart, lung, uterus, skin, spleen, plasma, blood) was removed, rinsed in saline, gently blotted dry, and weighed. Approximately 200 mg tissue was solubilized overnight at 60° C in 500 μl hyamine hydroxide and diluted in 0.1 *N* HCl to a total volume of 5.5 ml. Aliquots of this were analyzed for ^{14}C by standard liquid scintillation techniques. Cocktail T scintillant (8 ml) and 25 μl glacial acetic acid (to reduce the chemiluminiscence of hyamine) were added to 0.2- to 1-ml hyamine-solubilized tissue digests. Aliquots (0.1–1.5 ml) of hyamine-solubilized liver (spiked with ^{14}C standard) were used to determine a colour quench correction curve. Samples were counted by the method of external standardization; the counting efficiency was > 82%. Platinum analysis was carried out by flameless atomic absorption spectrophotometry as previously described by Siddik et al. [21].

Platinum and ^{14}C concentrations are expressed as nmol/g in tissue or nmol/ml in fluid. These respective concentrations were used to calculate the ^{14}C:Pt ratios in the tissues and fluids. Thus, a ratio of 1 in tissue indicated that carboplatin was probably in its intact form, as in the dose solution. Similarly, a ratio of < 1 would indicate a greater loss of ^{14}C than of Pt from the tissue.

Metabolic studies. Mice injected i.v. with ^{14}C-carboplatin (80 mg/kg; 1.1 mCi/kg) or ^{14}C-cyclobutane dicarboxylic acid (31 mg/kg; 1.1 mCi/kg) were housed in glass Metabowls (Jencons Scientific, Leighton Buzzard, UK) for 48 h. Exhaled CO_2 was trapped in 1-ethanolamine:2-methoxyethanol (1:2) and samples were analyzed for ^{14}C at 6 h and at 1 and 2 days posttreatment as described by Siddik et al. [19]. Flameless atomic absorption spectrophotometry was used to determine Pt levels in urine and in faeces digested in concentrated nitric acid. For faecal ^{14}C analysis, samples were oxidized using a Packard oxidizer (recovery > 98%) prior to counting by liquid scintillation.

Pharmacokinetic analysis. The plasma or tissue concentration-time data for both Pt and ^{14}C were analyzed by a two-compartment, open model using a nonlinear least squares computer program [18], as described by the equation:

$$C_o = A_{exp}{}^{(-at)} + B_{exp}{}^{(-\beta t)}).$$

Results

All tissues of mice receiving ^{14}C-carboplatin (80 mg/kg; 1.1 mCi/kg) showed a biphasic decline in both ^{14}C and Pt levels over 5 days, as exemplified by the plasma, liver, and kidney in Fig. 2. At 5 min post treatment, Pt concentrations in the plasma, liver, and kidney were 446, 138, and 1405 nmol Pt/ml or per gram (Table 1). These concentrations fell rapidly with time. The initial rate of loss of Pt and ^{14}C from all tissues (over the 2 h) was similar. Thereafter, the rate of removal of ^{14}C exceeded the rate of Pt removal (Table 2). In the liver, the pattern of ^{14}C removal differed from that in all other tissue, since the ^{14}C curve levelled out in the terminal phase.

The distribution of ^{14}C-cyclobutane dicarboxylic acid (31 mg/kg; 1.1 mCi/kg) was studied to compare the tissue distribution and elimination phase half-lifes with those of ^{14}C-carboplatin. The highest ^{14}C-cyclobutane dicarboxylic acid levels were observed in the kidney (Table 3) and may be due to the high urinary excretion of this species. The half-lifes in plasma and tissue for the two compounds differed (Table 4). Interestingly, there was no substantial dif-

Table 1. Platinum concentrations (nmol/g) in tissues of mice receiving ^{14}C-Carboplatin (80 mg/kg; 1.1 mCi/kg)

Tissue	5 min	1 h	4 h	8 h	1 day	2 days	5 days
Liver	138± 26.1	108 ±20.5	34.9±2.0	28.8±3.1	26.7± 0.3	19.8±2.6	13.2±1.6
Kidney	1405±107	158 ±92.3	46.1±4.6	30.8±7.8	37.4±18.5	26.8±6.15	12.3±2.6
Muscle	77± 8.3	17.1± 4.1	6.6±0.4	6.9±0.6	2.6± 0.1	3.2±0.3	1.7±0.1
Ileum	66± 5.1	45.7± 6.1	19.8±3.1	15.7±2.6	7.2± 1.1	4.5±1.0	2.3±0.5
Heart	117± 16.1	16.5± 1.9	6.5±0.6	4.4±0.4	3.2± 0.2	2.9±0.5	2.3±0.6
Lung	241± 17.3	35.9± 1.9	16.0±1.6	13.7±1.4	6.6± 0.3	5.8±0.8	4.1±0.3
Uterus	287± 23.1	33.6± 2.6	16.6±2.2	9.4±1.3	7.3± 0.6	5.1±0.1	12.9±2.4
Skin	246± 42.0	36.9± 3.3	16.5±2.1	16.4±1.5	8.9± 1.1	10.4±0.5	9.3±0.3
Spleen	77± 7.8	38.3± 5.5	15.0±1.8	18.0±2.0	16.8± 2.0	18.4±1.0	18.7±2.5
Plasma	446±201	45.7± 4.6	4.9±0.3	3.9±0.1	2.5± 0.2	1.7±0.1	0.6±0.05
Blood	195± 88.1	83.0±11.3	24.5±4.2	16.0±1.8	6.8± 0.6	6.5±0.6	6.9±0.51

Results are expressed as $x \pm$ SEM, $N = 4$

Table 2. ^{14}C concentrations (nmol/g) in tissues of mice receiving ^{14}C-carboplatin (80 mg/kg; 1.1 mCi/kg)

Tissue	5 min	1 h	4 h	8 h	1 day	2 days	5 days
Liver	150 ± 13.3	102 ±50.3	18.8±0.9	10.6±0.5	7.5±0.5	5.9±0.2	5.8±0.2
Kidney	1555 ±112	171 ±37	26.3±1.7	14.3±0.6	5.8±0.9	1.8±0.1	0.5±0.01
Muscle	59 ± 6.3	8.2± 0.4	3.4±0.2	2.9±0.2	0.9±0.2	0.5±0.05	0.2±0.03
Ileum	136 ± 7.8	48.0± 2.8	11.9±0.3	6.5±0.4	1.8±0.4	0.6±0.07	0.2±0.07
Heart	91.4± 10.1	14.3± 1.5	4.3±0.1	2.7±0.1	0.9±0.07	0.5±0.05	0.2±0.03
Lung	244 ± 21.3	39.8± 3.6	13.9±1.2	9.2±0.5	2.3±0.2	0.9±0.07	0.2±0.02
Uterus	289 ± 13.8	40.5± 5.9	15.0±0.9	6.9±0.4	1.8±0.4	0.9±0.1	0.3±0.02
Skin	194 ± 40.9	26.9± 1.8	10.4±0.1	5.8±0.3	1.9±0.1	1.0±0.05	0.3±0.01
Spleen	71 ± 8.6	33.8± 0.2	13.1±0.9	10.2±0.9	6.3±1.0	5.2±0.5	3.3±0.1
Plasma*	691 ±181	46.5± 7.0	4.1±0.07	2.7±0.5	1.1±0.2	0.3±0.01	<0.1
Blood*	412 ± 93.1	52.3± 1.6	6.8±0.3	4.5±0.08	1.8±0.08	0.6±0.01	<0.1

Results are expressed as $x \pm$ SEM, $N = 4$

* Plasma and blood ^{14}C concentrations at 5 days were <0.1 nmol/ml and have been excluded from calculations of terminal half-lives

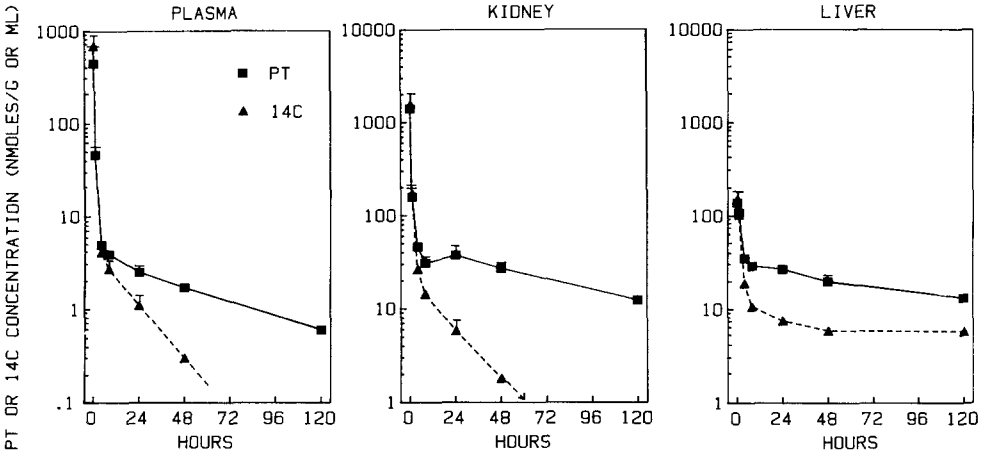

Fig. 2. Decay of Pt (■) and ^{14}C (▲) in plasma, kidney, and liver following ^{14}C-carboplatin (80 mg/kg; 1.1 mCi/kg; i.v.) over 5 days. Results are expressed as mean ± SEM, $N = 4$

ference in the retention or elimination half-lifes of ^{14}C-cyclobutane dicarboxylic acid in liver compared with that in other tissues (Tables 3 and 4).

The elimination phase half-lifes for Pt varied between different types of tissue, from 40 h in plasma to 156 h in skin (Table 4). The corresponding half-lifes for ^{14}C were similar for most tissue (18–35 h), although the liver and spleen were exceptions (210 and 90 h, respectively).

The temporal changes in the normalized ^{14}C:Pt ratios are illustrated for four types of tissue in Fig. 3. A ratio of unity was demonstrated at 1 h and maintained for up to 2 h in all tissues. In most types of tissue the ratio decayed rapidly thereafter, whereas in the liver the ratio at 5 days was 2- to 10-fold greater than in other tissues. At 5 days, the normalized ^{14}C:Pt ratios in most tissues were 0.04–0.24. In the liver, on the other hand, only 48% of the

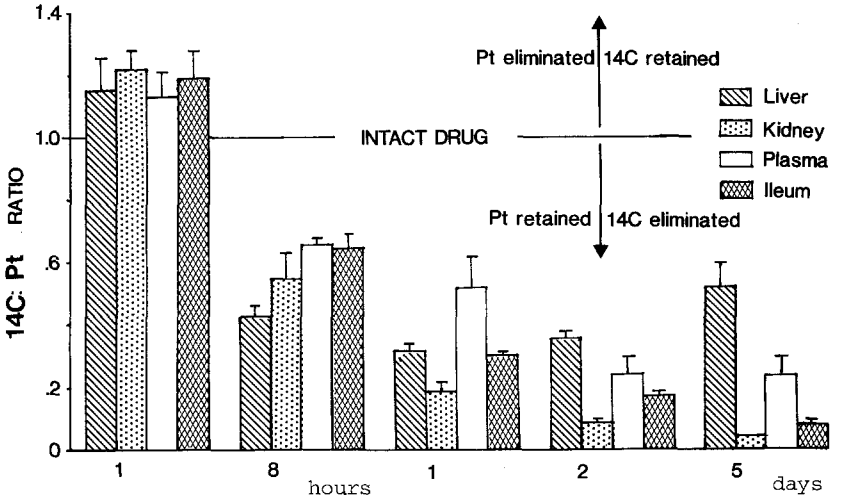

Fig. 3. Tissue ^{14}C:Pt ratios in four tissues. Intact carboplatin is represented by a ratio of unity. Results are expressed as mean ± SEM, $N = 4$

Table 3. ^{14}C concentrations (nmol/g) in tissues of mice receiving 1,1-^{14}C-cyclobutane dicarboxylic acid

Tissue	5 min	1 h	4 h	8 h	1 day	2 days	5 days
Liver	1078 ±333	29.1± 8.9	3.7±0.4	2.0±0.2	1.7±0.06	1.4±0.02	1.0±0.01
Kidney	1235 ±197	380 ±166	14.9±4.5	7.2±0.3	5.5±0.6	4.8±0.4	2.4±0.07
Muscle	66.9± 4.9	13.8± 3.9	1.2±0.3	0.2±0.01	0.2±0.02	0.1±0.01	0.1±0.01
Ileum	58.5± 12.5	21.0± 5.9	3.3±0.6	0.9±0.07	0.6±0.03	0.5±0.02	0.2±0.01
Heart	110 ± 3.8	25.1± 7.26	0.9±0.3	0.3±0.01	0.3±0.03	0.3±0.03	0.2±0.01
Lung	216 ± 10	56.7± 20	2.9±3.1	0.7±0.07	0.3±0.02	0.3±0.01	0.1±0.01
Uterus	140 ± 29	44.3± 11.4	10.7±3.1	0.9±0.07	1.2±0.1	0.5±0.09	0.2±0.05
Skin	144 ± 4.9	25.4± 8.9	0.9±0.4	0.4±0.02	0.4±0.04	0.8±0.2	0.3±0.01
Spleen	71 ± 2.4	25.5± 7.3	4.2±0.9	1.4±0.2	0.6±0.04	0.6±0.03	0.3±0.01
Plasma*	412 ±129	241 ±146	2.2±0.8	0.3±0.01	0.1±0.01	0.1±0.01	<0.1
Blood*	388 ± 14.5	64.8± 21.0	1.9±0.9	0.7±0.2	0.3±0.1	0.3±0.1	<0.1

Results are expressed as $x \pm$ SEM, $N = 4$

* Plasma and blood ^{14}C concentrations at 5 days are <0.1 nmol/ml and have been excluded from calculations of terminal half-lives

Table 4. Elimination half-lives of platinum and ^{14}C in tissues of mice receiving ^{14}C-carboplatin or ^{14}C-cyclobutane dicarboxylic acid

Tissue	Half-life (h)		
	Carboplatin		^{14}C-CBDCA
	PT	^{14}C	
Plasma	39.8± 6.0	27.2± 8.6	54.7±25.8
Liver	98.9± 39.0	210.2±127§	105.3±46.6
Kidney	70.7± 23.6	25.8± 6.4	74.1±26.7
Ileum	54.1± 19.4	25.9± 7.6	71.1±26.2
Muscle	116.4± 83.0	31.5± 10.2	150.8±72.3
Heart	43.5± 10.8	27.1± 8.2	97.2±73.3
Spleen[a]	–	90.3± 30.0[b]	78.4±35.8
Skin[a]	156.4±100.6	28.8± 4.4	–
Blood	145.8± 98.4	17.6± 1.6	27.5±16.8
Uterus[a]	–	34.8± 7.0	83.7±30.8
Lung	143.2±100.0	31.5± 5.6	70.2±33.0

Results are expressed as $x \pm$ SD; $N = 4$

[a] Half-lives could not be determined due to absence of a well-defined decay in these tissues over 2–120 h

[b] ^{14}C Half-lives significantly ($P \leq 0.01$) longer than in other tissues

carboplatin could be ascribed to transformed Pt species. The extent of transformation in this tissue on day 1, however, was about 65%–70%, which was similar to that in the kidney and other tissues.

Metabolic studies

Excretion of ^{14}C-carboplatin (80 mg/kg; 1.1 mCi/kg) or ^{14}C-cyclobutane dicarboxylic acid (31 mg/kg; 1.1 mCi/kg) was primarily renal, with 86%–92% of the carboplatin and 96% of the ^{14}C-cyclobutane dicarboxylic acid eliminated in the urine at 4 days (Table 5). Faecal excretion accounted for 1% and 4% of ^{14}C and Pt, respectively, of the administered dose of ^{14}C-carboplatin. Low faecal excretion of ^{14}C-cyclobutane dicarboxylic acid was also obtained (3.5% dose). A negligible amount of the administered dose of ^{14}C-carboplatin (0.06%) and of ^{14}C-cyclobutane dicarboxylic acid (0.6%) was exhaled as $^{14}CO_2$.

Discussion

The chemical activation of cisplatin and possibly of carboplatin, through an aquation step, is thought to be rate-limiting in the subsequent platination process [3, 10]. The

Table 5. Cumulative excretion of carboplatin (80 mg/kg; 1.1 mCi/kg) and cyclobutane dicarboxylate ligand (31 mg/kg; 1.1 mCi/kg) over 4 days in mice

Time (h)	Urinary excretion (% dose)			CO_2 excretion (% dose)	
	Carboplatin		^{14}C-CBDCA	Carboplatin	^{14}C-CBDCA
	Pt	^{14}C	^{14}C		
24	83.4 ± 2.7	88.1 ± 3.1	91.3 ± 4.9	0.049 ± 0.001	0.57 ± 0.03
48	85.1 ± 2.8	91.5 ± 4.0	93.8 ± 5.4	0.054 ± 0.001	0.60 ± 0.03
72	85.5 ± 2.8	91.8 ± 3.9	94.8 ± 4.8	0.057 ± 0.001	0.63 ± 0.03
96	85.6 ± 2.8	91.9 ± 4.1	95.7 ± 4.0	0.059 ± 0.01	0.65 ± 0.02

Results are expressed as $x \pm$ SE, $N = 4$

bidentate cyclobutane dicarboxylate ligand of carboplatin confers stability to the molecule and thereby renders carboplatin 10- to 20-fold less reactive than the parent, cisplatin [8, 20]. The rate of activation within a tissue may relate to its biochemical and chemical composition. In a previous study [20], we have demonstrated that binding of cisplatin and carboplatin to the liver and kidney significantly exceeded the rate of binding to plasma. Hence, through the synthesis of ^{14}C-carboplatin it has been possible to examine the activation of carboplatin in tissue and to trace the fate of the Pt and ^{14}C moieties.

The data in this communication indicate that carboplatin remains intact in plasma and tissue for up to 2 h. Thereafter the ligand is released from the molecule and preferentially removed from the tissue, as indicated by decreasing values for the ^{14}C:Pt ratios in tissue. The decay of both Pt and ^{14}C in all tissues is biphasic, with a rapid initial phase and a slow elimination phase. This is similar to the distribution pattern of Pt previously observed with cisplatin in rats and mice [6, 19]. Prolonged retention of platinum in tissue suggests irreversible or tight binding to tissue components. At 5 days posttreatment, Pt concentrations were greatest in the spleen, kidney, liver, and whole blood. High values for Pt in these organs following treatment with cisplatin have also been observed in rats [6, 12] and man [15]. Splenic Pt levels were 1.5-fold greater than kidney levels at 5 days, due to a continual uptake of Pt by this organ. The degradation of accumulated cell products such as Pt-containing erythrocytes may partly account for this phenomenon [13, 14].

The half-lifes of Pt in tissue were highly variable (40–156 h), and may be due to varying rates of intrinsic repair processes and subsequent excision of the Pt lesion, together with varying rates of protein turnover at platination sites within tissue.

The activation of carboplatin would involve a Pt-O bond in nucleophilic displacement, resulting in the formation of a monodentate Pt species. Subsequent loss of the ^{14}C ligand occurs, with a half-life of around 27 h. Since the ^{14}C elimination half-lifes in tissue were similar, the activation of carboplatin occurs at a similar rate in most of the tissue examined. Knox et al. [8] have shown that, following the initial binding of ^{14}C-carboplatin to DNA, the ^{14}C-cyclobutane dicarboxylic acid ligand is released from the isolated DNA with a half-life of 14.5 h, with no corresponding loss of Pt. Thus, the loss of ^{14}C ligand occurs as a result of the fragmentation of the molecule and not through the loss of the bound drug. Of the remaining Pt in tissue, a maximum of 4%–24% exists as intact drug and the rest is probably irreversibly bound. This assumes that all the ^{14}C present in tissue at 5 days is intact carboplatin.

High hepatic ^{14}C levels may be due to monodentate binding or the incorporation of ^{14}C into endogenous compounds. In contrast to the liver, the kidney is an efficient tissue for ^{14}C ligand elimination. However, "activation" of the molecule was similar in all tissue examined, suggesting that an enzymatic activation may not be involved. There was minimal transformation of ^{14}C-carboplatin and ^{14}C-cyclobutane dicarboxylic acid to $^{14}CO_2$.

The parallel study using ^{14}C-cyclobutane dicarboxylic acid distribution did not clarify the fate of the ligand in carboplatin, since the ^{14}C in the two compounds was handled differently in vivo. The major route of excretion of carboplatin was via the kidney, with a small percentage of the dose excreted in faeces.

These results imply that the metabolic handling of carboplatin varies according to the tissue. The elimination of the ^{14}C-cyclobutane dicarboxylate species from tissues was similar. However, Pt elimination was generally slower and tissue-dependent.

References

1. Calvert AH, Harland SJ, Newell DR, Siddik ZH, Jones AC, McElwain TJ, Raju S, Wiltshaw E, Smith IE, Baker JW, Peckham MJ, Harrap KR (1982) Early clinical studies with cis-diammine-1,1-cyclobutane dicarboxylate platinum II. Cancer Chemother Pharmacol 9: 140
2. Cleare MJ (1974) Transition metal complexes in cancer chemotherapy. Coord Chem Rev 12: 349
3. Cleare MJ, Hydes PC, Malerbi BW, Watkins DM (1978) Antitumour platinum complexes: relationships between chemical properties and activity. Biochimie 60: 835
4. Foster BJ, Clagett-Carr K, Leyland-Jones B, Hoth D (1985) Results of NCI-sponsored phase I trials with carboplatin. Cancer Treat Rev 12 [Suppl A]: 43
5. Harrap KR, Jones M, Wilkinson CR, Clink HMcD, Sparrow S, Mitchley B, Clarke S, Veasy A (1980) Antitumour, toxic and biochemical properties of cisplatin and eight other platinum complexes. In: Cisplatin: Current status and new developments. Academic Press, New York, p 193
6. Harrison R, McAuliffe LA, Zaki A, Brer J, Sharma H, Smith A, Jackson H, Fox BW (1983) A comparative study of the distribution in the male rat of platinum-labelled cis-dichlorodiammine platinum (II), cis-dichloro-bis-cyclopropylamine platinum (II). Cancer Chemother Pharmacol 10: 90
7. Johnson Matthey Patent Specification 1380228. Appl. No. 27304/73

8. Knox RJ, Friedlos F, Lydall DA, Roberts JJ (1986) Mechanism of cytotoxicity of anticancer platinum drugs: evidence that cis-diamminedichloro platinum (II) and cis-diammine (1,1-cyclobutane dicarboxylo)platinum (II) differ only in the kinetics of their interaction with DNA. Cancer Res 46: 1972
9. Krakoff IH (1979) Nephrotoxicity of cisdichlorodiammine platinum (II). Cancer Treat Rep 63: 1527
10. Leroy FA, Lutz R, Dedrick F, Litterst C, Guarino A (1979) Pharmacokinetic study of cis-dichloroplatinum (II) (DDP) in the Beagle dog. Thermodynamic and kinetic behaviour of cisplatin in a biological medium. Cancer Treat Rep 63: 59
11. Levine BS, Henry MC, Port CP, Richter WR, Urbanek MA (1981) Nephrotoxic potential of cis-diamminedichloroplatinum and four analogues in male Fischer F344 rats. J Natl Cancer Inst 67: 201
12. Litterst CL, Leroy AF, Guarino AM (1979) Disposition and distribution of platinum following parenteral administration of cis-dichlorodiammine platinum (II) to animals. Cancer Treat Rep 63: 1485
13. Long DF, Patton TF, Repta AJ (1981) Platinum levels in human erythrocytes following intravenous administration of cisplatin: Importance of erythrocytes as a distribution site for platinum species. Biopharm Drug Dispos 2: 137
14. Manaka RC, Wolf W (1978) Distribution of cisplatin in blood. Chem-Biol Interact 22: 353
15. Owens SE, Thatcher N, Sharma H, Adam N, Harrison R, Smith A, Zaki A, Baer JC, McAuliffe CA, Crowther D, Fox BW (1985) In vivo distribution of radioactively labelled platinum complexes; cisdichlorodiammine platinum (II), cis-trans-dichlorodihydroxy-bis-isopropylamine)platinum (IV) and cis-diammine (1,1-cyclobutanedicarboxylate)platinum (II) in patients with malignant disease, using a gamma counter. Cancer Chemother Pharmacol 14: 253
16. Peckham MJ, Horwich A, Brada M, Drury A, Hendry WF (1983) Cisdiammine-1,1-cyclobutane dicarboxylate platinum II (carboplatin) in the treatment of testicular germ-cell tumours. A preliminary report. Cancer Treat Rev 12 [Suppl A]: 101
17. Prestayko AW, D'Aoust JC, Issell BF, Crooke ST (1979) Cisplatin (cis-diamminedichloroplatinum II). Cancer Treat Rev 6: 17
18. Sampson J (1970) Non linear least squares programme BMDX85. In: Dixon WJ (ed) BMD biomedical computer programs, X-series supplement. University of California Press, Berkeley, p 177
19. Siddik ZH, Barnes RD, Dring LG, Smith RL, Williams RT (1979) The fate of lysergic acid di[^{14}C]ethylamide ([^{14}C]LSD) in the rat, guinea pig and rhesus monkey and of [^{14}C]isoLSD in rat. Biochem Pharmacol 28: 3093
20. Siddik ZH, Dible SE, Boxall FE, Harrap KR (1986a) Renal pharmacokinetics and toxicity of cisplatin and carboplatin in animlas. In: McBrien DC, Slater TH (eds) Biochemical mechanisms of platinum antitumour drugs. IRL Press, Oxford, p 171
21. Siddik ZH, Boxall FE, Harrap KR (1986b) Tissue solubilisation in hyamine hydroxide for the flameless atomic absorption spectrophotometric determination of platinum. In: McBrien DCH, Slater TF (eds) Biochemical mechanisms of platinum antitumour drugs. IRL Press, Oxford, p 355
22. Smith IE, Evans BD (1985) Carboplatin (JM8) as a single agent and in combination in the treatment of small cell lung cancer. Cancer Treat Rev 12 [Suppl A]: 73
23. Wiltshaw E (1985) Ovarian trials at the Royal Marsden. Cancer Treat Rev 12 [Suppl A]: 67

Received August 10, 1987/Accepted January 27, 1988