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The disposition and kinetics of intravenous N-acetylcysteine in patients with paracetamol overdosage

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Summary. Seventeen patients received standard treatment with intravenous N-acetylcysteine for 18 episodes of severe poisoning with paracetamol (acetaminophen). The dose of N-acetylcysteine was 150 mg/kg given in 15 min followed by 50 mg/kg in 4 h and 100 mg/kg over the next 16 h. Liver damage was absent or mild on 13 occasions (ALT < 500 μ/l) and severe on 5 (ALT > 1000 μ/l).

Total plasma N-acetylcysteine was estimated by HPLC. The mean maximum plasma concentration after the initial loading dose was 554 mg/l. Concentrations then fell rapidly and after 12 h a mean steady-state level of about 35 mg/l was maintained. When the infusion was discontinued N-acetylcysteine disappeared with a half-life of 5.7 h. The mean steady-state volume of distribution, AUC, mean residence time and total clearance were 536 ml/kg, 1748 $mg \cdot h \cdot l^{-1}$, 2.91 h and 3.18 $ml \cdot min^{-1} \cdot kg^{-1}$. These values are generally consistent with those previously reported with much smaller doses and the disposition of N-acetylcysteine does not appear to be dose-dependent. The elimination of N-acetylcysteine was not impaired in the patients with severe liver damage, and the pharmacokinetic variables and plasma concentrations were similar in patients with and without hepatotoxicity.

The dosage schedule for intravenous N-acetylcysteine should probably be modified since adverse reactions invariably occur early when plasma concentrations are at their highest, and liver damage was prevented just as effectively at the lowest as at the highest C_{max}. High initial concentrations of N-acetylcysteine can be avoided with simple alternative regimens based on the kinetic data of this study.

Key words: N-acetylcysteine, paracetamol; acetaminophen overdose, pharmacokinetics, liver damage, adverse reactions, dose modifications

N-acetylcysteine has long been used as a mucolytic agent for the treatment of pulmonary disease and cystic fibrosis. Recent new indications include the prevention of hepatic necrosis following paracetamol overdosage [1, 2], protection against the toxicity of metals and alkylating agents [3] and reversal of acquired tolerance to the cardiovascular effects of organic nitrates [4]. N-acetylcysteine has chelating and nucleophilic properties, and its efficacy as a specific antidote for paracetamol poisoning depends primarily on its ability to stimulate glutathione synthesis [5].

The results of previous studies of the kinetics of N-acetylcysteine in man have varied depending on the analytical methods used, dose, formulation and route of administration. Absorption is rapid following oral administration of single doses of 100 to 600 mg, but the bioavailability is very low. Twenty to 30% of an intravenous dose is excreted unchanged in the urine and the mean elimination half-life has varied from less than 2 to more than 6 h in different reports [6–10]. Interpretation of analytical data is complicated because of the variety of forms in which N-acetylcysteine can exist. It can be present free or bound to proteins in the reduced or oxidised state, and it forms mixed disulphides with other thiols and SH groups of proteins [8, 9]. Total N-acetylcysteine may be estimated after regeneration of the reduced form with agents such as dithiothreitol, but the results obviously do not reflect its true disposition in vivo.

In the treatment of paracetamol (acetaminophen) overdosage, N-acetylcysteine is given intravenously in doses which greatly exceed those used previously for other indications and its disposition in such circumstances is unknown. In addition, it might be anticipated that its elimination would be impaired in patients who are treated late and suffer severe hepatic injury. The present study was therefore undertaken to investigate the disposition and kinetics of high dose intravenous N-acetylcysteine in patients with and without liver damage following paracetamol overdosage.

Table 1. Clinical details

Patient	Age (years) and sex	Plasma paracetamol (mg/l)	Time after ingestion (h)	Ingestion-treatment interval (h)	Other drugs taken[a]	Max. ALT (u/l)	Max. bilirubin (μmol/l)	Max. prothrombin time ratio	Plasma paracetamol half-life (h)
LT	22 F	231	4	5.5	E, M	13	9	1.2	3.6
LA	44 F	162	7.6	9.5	–	16	12	–	2.63
TG	13 F	237	4	5.5	–	16	4	1.1	2.71
JA	19 F	208	4	5.75	D, C, E	17	5	1.0	2.09
MC	21 F	189	6	6	–	19	8	1.2	1.67
JM	32 M	185	7.25	7.75	E	22	13	1.0	2.11
GM	20 M	143	11.2	13.5	P	28	7	1.0	1.92
JP	68 M	381	10.7	13	E	31	15	1.6	7.13
PF	33 M	361	4	7	E, P	45	11	1.1	2.96
KM	20 M	272	4.7	6	E	52	20	1.2	3.49
MT	38 F	600	2.25	3.5	E, K	375	16	1.4	6.37
EA	36 F	219	4	6	–	436	34	1.2	2.97
TM	20 M	97	8.7	10.8	P	488	13	1.3	2.81
MH(1)	31 F	95	8.75	8.75	–	1,950	59	1.6	4.86
MH(2)	31 F	185	5.2	8.25	–	1,560	46	2.8	4.4
HM	28 F	36	15.5	15.5	–	3,622	21	1.9	2.46
MW	15 F	50	11	14.5	H, A	6,650	23	2.2	3.39
GB	28 F	125	4	17	–	8,300	113	6.4	7.75

[a] E = Ethanol, A = Amoxycillin + clavulinic acid, C = Codeine, D = Doxylamine, H = Dihydrocodeine, K = Cephalexin, M = Ampicillin, P = d-Propoxyphene

Patients and methods

Patients and management

The study group consisted of 17 patients admitted after overdosage of paracetamol on 18 occasions. Their mean age was 28.7 years (range 13 to 68 years) and 6 patients were male. They were all admitted within 20 h of the overdose and were severely poisoned with plasma paracetamol concentrations above the "treatment line" joining semilogarithmic plots of 200 mg/l at 4 h after ingestion and 30 mg/l at 15 h [1]. Seven patients took other drugs in addition to the paracetamol and 5 also took ethanol. Clinical and laboratory details are summarized in Table 1.

Routine management consisted of gastric lavage or induction of emesis with syrup of ipecac in patients admitted within 4 h, administration of naloxone for suspected poisoning with narcotic analgesics, and maintenance intravenous fluids together with the cautious use of metoclopramide for persistent vomiting. Treatment with intravenous N-acetylcysteine (Parvolex, Duncan, Flockhart & Co. Ltd.) was started within 8 to 10 h when indicated by the plasma paracetamol concentration on admission or as soon as possible in patients admitted after this time. Twelve patients were treated within 10 h, 4 between 10 and 15 h and 2 after 15 h. In Patient GB the plasma paracetamol concentration was only 124 mg/l on admission at 4 h but she became unwell later and N-acetylcysteine was started when it was found that the plasma paracetamol had risen above the treatment line. The N-acetylcysteine was infused by pump in an initial dose of 150 $mg \cdot kg^{-1}$ in 200 ml of 5% dextrose over 15 min followed by 50 $mg \cdot kg^{-1}$ in 500 ml of 5% dextrose in 4 h and 100 $mg \cdot kg^{-1}$ in a litre of 5% dextrose over the next 16 h. The total dose was 300 $mg \cdot kg^{-1}$ given over 20.25 h [1] and the mean total dose for the whole group was 18.07 g.

Venous blood was sampled at 0, 0.25, 0.5, 0.75, 1.25, 2.25, 4.25, 8.25, 12.25, 16.25, 20.25, 21.25, 22.25, 24.25, 28.25, and 36 h after starting treatment for estimation of plasma N-acetylcysteine and paracetamol. Blood was also taken twice daily for at least 3 days for liver function tests and measurement of plasma electrolytes, urea and creatinine. Informed consent was obtained and the study was approved by the local ethics committee.

Analytical methods

Blood for the assay of N-acetylcysteine was taken in lithium heparin tubes, refrigerated immediately, centrifuged within 8 h and the plasma stored at −20° C. With this technique there was no loss of total N-acetylcysteine content compared with immediate assay. N-Acetylcysteine was estimated by the high performance liquid chromatographic method of Lewis et al. [11] with modifications similar to those described subsequently [12, 13]. Plasma was diluted with water as required, and N-glycylglycine pre-reacted with 2,4-dinitrofluorobenzene was used as the internal standard. The ultrafiltration step was omitted and after reaction with dinitrofluorobenzene, the mixture was acidified with 0.5 ml of 1 M HCl before extraction into ether. The glycylglycine and N-acetylcysteine derivatives eluted in 2.8 and 5.3 min respectively. The method measures total oxidised and reduced N-acetylcysteine together with drug bound to other thiols and proteins.

Paracetamol in plasma was estimated by high performance liquid chromatography [14], and standard laboratory methods were used for clinical biochemical measurements.

Pharmacokinetic and statistical analysis

The plasma concentration-time data for N-acetylcysteine were fitted to an open 2 compartment model by weighted non-linear regression analysis using the SIPHAR[1] pharmacokinetic curve fitting and modelling programme. In addition to the coefficients of the model, this provided estimates of the area under the plasma concentration-time curve (AUC), total body clearance (dose/AUC), mean residence time (MRT), steady state volume of distribution (V_Z) (clearance × MRT) and the terminal plasma half-life. Predicted plasma concentrations of N-acetylcysteine for different infusion regimens were modelled with the SIPHAR programme using the mean coefficients and exponents for the whole group of patients. The plasma paracetamol half-life was calculated by weighted linear regression from data obtained during the first 8 h of treatment with N-acetylcysteine.

Results are expressed as means (SD) and the statistical significance of differences between means was determined by the Mann-Whitney test taking $p = < 0.05$ as the level of significance.

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Results

All the patients completed the course of treatment without incident. Three patients had mild liver damage with maximum plasma alanine aminotransferase (ALT) activity less than 500 $U \cdot l^{-1}$ and in 5 liver damage was severe with ALT activity exceeding 1000 $U \cdot l^{-1}$. All the patients recovered and liver function tests returned to normal within 7 to 14 days. Comparisons were made between the 13 patients with mild or no liver damage and the 5 with severe liver damage.

Plasma concentrations of N-acetylcysteine

The mean maximum plasma concentration of total N-acetylcysteine (C_{max}) in the whole group of patients after administration of the initial loading dose was 554 (129) mg/l (range 304 to 875 ml/l). Concentrations fell progressively for about 12 h and then remained relatively constant (Fig. 1). The mean "steady-state" plasma concentration at the end of the infusion was 33.4 (16.7) mg/l but there was considerable individual variation and concentrations ranged from 11 to 90 mg/l. When the infusion was discontinued N-acetylcysteine disappeared rapidly from the plasma with a mean half-life of 5.7 (2.9) h.

The mean plasma concentrations of N-acetylcysteine were actually lower in the patients with severe liver damage than those without, but the differences between the groups were not significant at any time (Fig. 2).

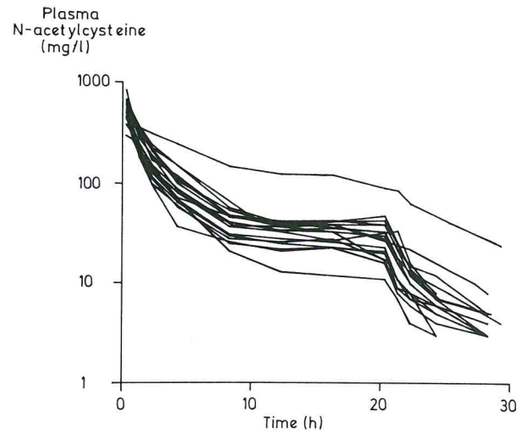

Fig. 1. Plasma concentrations of total N-acetylcysteine following infusion of 150 $mg \cdot kg^{-1}$ in 15 min, 50 $mg \cdot kg^{-1}$ in 4 h and 100 $mg \cdot kg^{-1}$ in 16 h in 18 patients with paracetamol overdosage

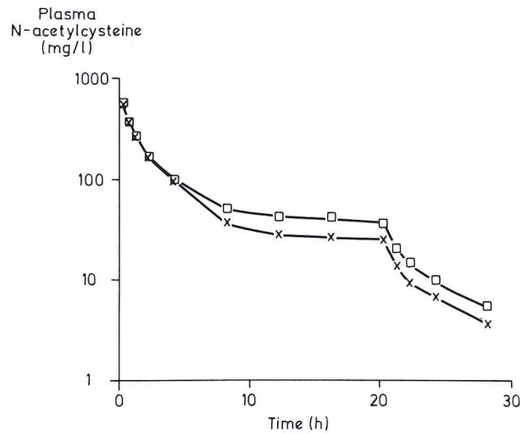

Fig. 2. Mean plasma concentrations of total N-acetylcysteine in 13 patients with mild or no liver damage (□) and 5 with severe liver damage (×) following paracetamol overdosage

Kinetic analysis

Good fits of the data to the two compartment model were obtained with all patients except GB in whom the best fit was with a single compartment. The mean values of the coefficients and exponents A, B, α and β for the whole group were 1120 (264) and 104 (123) mg/l, and 1.0 (0.23) and 0.15 (0.08) h respectively ($n = 17$). The values were similar in the patients who had severe liver damage and those who did not.

The pharmacokinetic variables for the two groups of patients are summarised in Table 2. Overall, the mean values for the steady-state volume of distribution, AUC, mean residence time and total body clearance of N-acetylcysteine were 536 (210) $ml \cdot kg^{-1}$, 1748 (695) $mg \cdot l^{-1} \cdot h$, 2.91 (1.0) h and 3.18 (0.93) $ml \cdot min^{-1} \cdot kg^{-1}$ respectively. There was considerable individual variation, and the terminal half-life and clearance ranged from 1.23 to 10.46 h and 1.21 to 5.14 $ml \cdot min^{-1} \cdot kg^{-1}$ respectively. There were no significant differences in any of the pharma-

Table 2. Pharmacokinetic variables for intravenous N-acetylcysteine in patients with and without severe liver damage following paracetamol overdosage

Patients	Total dose of N-acetylcysteine (g)	C_{max} ($mg \cdot l^{-1}$)	Steady state plasma concentration ($mg \cdot l^{-1}$)	Plasma half life (h)	Volume of distribution (Vz) ($ml \cdot kg^{-1}$)	AUC ($mg \cdot l^{-1} \cdot h$)	Mean residence time (h)	Total body clearance ($ml \cdot min^{-1} \cdot kg^{-1}$)
Without severe liver damage ($n=13$)	17.6 (4.30)	538 (104)	36.7 (17.6)	5.52 (2.52)	526 (205)	1805 (755)	2.97 (1.09)	3.11 (0.92)
With severe liver damage ($n=5$)	19.2 (7.22)	596 (170)	24.8 (10.0)	6.06 (3.64)	562 (221)	1598 (476)	2.75 (0.72)	3.39 (0.90)

cokinetic variables in the patients with and without liver damage of any severity. In particular, the mean clearance of N-acetylcysteine was not reduced and its plasma half-life was not prolonged in the patients with severe liver damage.

The findings were atypical in patient JP who had the highest plasma concentrations of N-acetylcysteine (Fig. 1) with the greatest AUC (4126 $mg \cdot l^{-1} \cdot h$) and the lowest clearance (1.21 $ml \cdot min^{-1} \cdot kg^{-1}$). The plasma paracetamol half-life was also greatly prolonged at 7.13 h but there was no evidence of acute liver damage.

Correlations with liver function

There were no significant correlations between the half-life, clearance, AUC, C_{max} and "steady-state" plasma concentration of N-acetylcysteine, and maximum plasma ALT activity, bilirubin concentration or prothrombin time ratio. The plasma paracetamol half-life was not related to any of the pharmacokinetic variables for N-acetylcysteine except for a weak but significant correlation with the AUC ($r=0.53$, $p=<0.05$). There were significant correlations between the paracetamol half-life and the maximum plasma bilirubin and prothrombin time ratio ($r=0.65$, $p=<0.01$), but not the ALT activity. As expected, the plasma ALT, bilirubin and prothrombin time ratio were all significantly correlated with each other ($r=0.74$ to 0.90, $p=<0.001$).

Predicted plasma N-acetylcysteine concentrations with different infusion rates

Examples of the patterns of plasma concentrations of N-acetylcysteine which could theoretically be produced by simple combinations of different infusion rates are shown in Fig. 3. The simulations were based on the mean coefficients and exponents derived from the compartmental analysis assuming administration of the standard total dose of 300 $mg \cdot kg^{-1}$ in 20.3 h.

With constant input of 70 $mg \cdot kg^{-1}$ in 20 min, 100 $mg \cdot kg^{-1}$ in 2 h, and 130 $mg \cdot kg^{-1}$ in 18 h there would be an initial plateau concentration of about 280 $mg \cdot l^{-1}$ for 2 h followed by a rapid fall to a level of about 50 $mg \cdot l^{-1}$ at 8 h which is maintained until the end of the infusion (Fig. 3 a). A similar pattern with the initial plateau held at 150 $mg \cdot l^{-1}$ for 6 h and a final steady-state level of about 50 $mg \cdot l^{-1}$ could be obtained by sequential infusion of 40 $mg \cdot kg^{-1}$ in 20 min, 80 $mg \cdot kg^{-1}$ in 3 h, 75 $mg \cdot kg^{-1}$ in 3 h and 105 $mg \cdot kg^{-1}$ in 14 h (Fig. 3 b).

Discussion

The standard intravenous dosage regimen produced very high initial plasma concentrations of total N-acetylcysteine which ranged from 304 to 875 mg/l. Concentrations subsequently fell rapidly and after about 12 h a relatively constant level of about 35 mg/l was maintained until the infusion was discontinued. N-acetylcysteine then disappeared with a mean terminal half-life of about 6 h. It had a relatively small volume of distribution of 536 $ml \cdot kg^{-1}$ and the mean clearance was 3.18 $ml \cdot min^{-1} \cdot kg^{-1}$ (equivalent to 37.5 l and about 225 $ml \cdot min^{-1}$ respectively in a 70-kg patient). These results may be compared with those of studies in healthy volunteers given doses of 200 to 600 mg. Olsson et al. [8] reported a mean volume of distribution, clearance and half-life of total N-acetylcysteine of 470 $ml \cdot kg^{-1}$, 1.83 $ml \cdot min^{-1} \cdot kg^{-1}$ and about 6 h respectively, while the corresponding values in another study were 330 $ml \cdot kg^{-1}$, 3.5 $ml \cdot min^{-1} \cdot kg^{-1}$ and 2.27 h [7]. Despite the discrepancies, the results in the present study were of the same general order, and the kinetics of total N-acetylcysteine do not seem to be dose-dependent. The reduced form is normally cleared very rapidly [8], and unfortunately it could not be measured under the emergency conditions of the present study. N-Acetylcysteine probably acts indirectly by releasing free cysteine [9], and the concentration of total drug may not be a reliable index of its biological action.

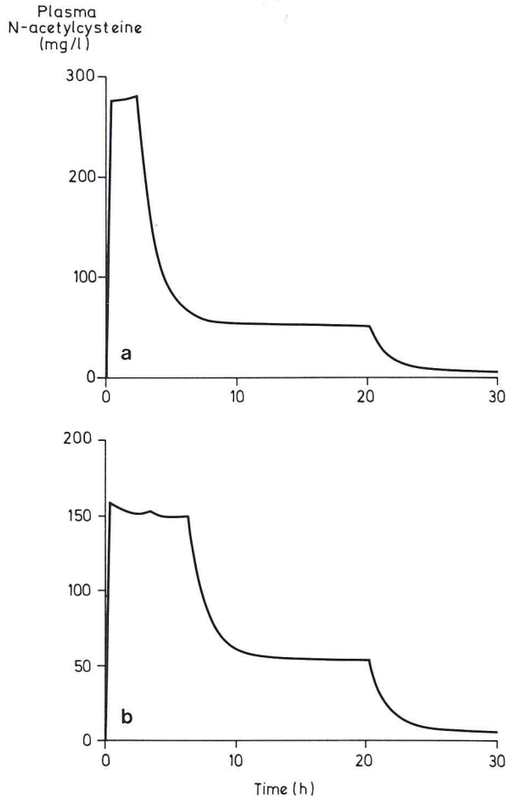

Fig. 3 a, b. Predicted plasma concentrations of total N-acetylcysteine according to the mean coefficients and exponents obtained by 2 compartmental analysis of data from 17 patients. **a** Zero order infusion was assumed at a rate of 70 $mg \cdot kg^{-1}$ for 20 min, 100 $mg \cdot kg^{-1}$ for 2 h and 130 $mg \cdot kg^{-1}$ for 16 h. **b** Constant infusion was assumed at a rate of 40 $mg \cdot kg^{-1}$ for 20 min, 80 $mg \cdot kg^{-1}$ for 3 h, 75 $mg \cdot kg^{-1}$ for 3 h and 105 $mg \cdot kg^{-1}$ for 14 h

There was considerable individual pharmacokinetic variation in our patients, and the findings in patient JP were anomalous. He was found to have multiple pulmonary opacities on chest X-ray and a mass in the left kidney consistent with a diagnosis of hypernephroma but otherwise there was no obvious explanation for the very low clearances of N-acetylcysteine and paracetamol.

An unexpected finding was that the clearance of N-acetylcysteine was not reduced and its plasma concentrations were not increased in the patients with severe liver damage. Maximum impairment of hepatic function is delayed for at least 3 days after overdosage of paracetamol, and it is possible that the enzymes involved in the metabolism of N-acetylcysteine were still functioning normally at this early stage. On the other hand, the metabolism of paracetamol and other drugs is impaired from the outset in patients who develop severe liver damage [15] and in keeping with this, the paracetamol half-life in the present study was prolonged in relation to the severity of liver damage as shown by the plasma bilirubin and prothrombin time ratio. Other possible explanations for the normal clearance of N-acetylcysteine include greater utilisation in patients with toxicity and elimination from sites other than the liver. N-acetylcysteine is deacetylated in the liver in the rat [16], but it is also rapidly metabolised to cysteine and inorganic sulphite in the gastrointestinal tract [17]. Reservations have been expressed about the safety of late treatment with N-acetylcysteine in patients with extensive hepatic necrosis. Our observations are reassuring in this respect and it seems unlikely that there would be abnormally high concentrations of N-acetylcysteine in such circumstances.

The dosage regimen for intravenous N-acetylcysteine was originally chosen on an arbitrary basis [18]. Its efficacy was known to fall off rapidly when treatment was delayed beyond 8 to 10 h, and the object was to give the highest tolerable dose for a short period in the hope of reversing the events leading to hepatic necrosis at a critical stage. The dose was then reduced and the infusion discontinued after 20.25 h. This regimen has not been changed and although very effective, it is most unlikely to be optimal. It is not known whether the initial very high concentrations of N-acetylcysteine are necessary for full protection against hepatotoxicity, but the patients in the present study with the lowest C_{max} fared just as well as those with the highest. Perhaps more importantly, excessively high concentrations are likely to predispose to adverse reactions. Flushing, urticaria, hypotension and bronchospasm may occur rarely, and similar but more severe reactions have been reported after overdosage of N-acetylcysteine [19, 20]. Typically these "anaphylactoid" reactions occur shortly after starting treatment at a time when the concentrations of N-acetylcysteine are highest [21]. Thus these effects appear to be concentration-dependent and they are more likely to represent pharmacological effects than "Type B" adverse reactions.

The dosage of intravenous N-acetylcysteine should probably be modified now to avoid these high initial concentrations. Unfortunately, no information is available concerning the concentration-time-response relationships, or even the optimum duration of therapy. However, treatment may reasonably be continued for 24 h, and the infusion rates used to generate the predicted plasma concentrations in Fig. 3 may severe as a guide to the develop-

ment of a more rational dosage schedule. Irrespective of any changes, early treatment will still be the most critical factor for prevention of liver damage after paracetamol overdosage.

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Application of a radioreceptor assay in a pharmacokinetic study of oxitropium bromide in healthy volunteers after single i.v., oral and inhalation doses

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Summary. Oxitropium bromide (OXBR) is a new anticholinergic drug, which is expected to be useful in the treatment of nocturnal asthma. The only pharmacokinetic data were obtained with the ^{14}C-labelled compound. A sensitive radioreceptor assay for the determination of unlabelled OXBR in plasma was developed, based on competition between OXBR and ^{3}H-N-methylscopolamine for binding to muscarinic receptors. OXBR was isolated from plasma by ion-pair extraction and re-extraction. Active metabolites present in significant amounts might interfere in the assay, but this was not the case for OXBR metabolites. Detection limits were 300 $pg \cdot ml^{-1}$ and 3 $ng \cdot ml^{-1}$ for plasma and urine, respectively. For the latter no extraction step was required. The single dose pharmacokinetics of OXBR was studied following inhalation (3 mg), oral (2 mg) and i.v. (1 mg) administration to 12 men, following an open, cross-over design.

After i.v. administration the kinetic parameters were: V_c 38.4 l; $t_{1/2\alpha}$ 5.3 min; $t_{1/2\beta}$ 142 min; AUC 8.9 $h \cdot ng \cdot ml^{-1}$; renal excretion 50.2%, k_{10} 3.5 $l \cdot h^{-1}$ and total clearance 1874 ml/min. The apparent bioavailabilities were 0.48% and 12.4% by the oral and inhalation routes, respectively, based on the cumulative renal excretion. There were moderate adverse reactions due to the anticholinergic properties of the drug.

Key words: oxitropium bromide; pharmacokinetics, radioreceptor assay, side-effects

Oxitropium bromide (OXBR) is a new anticholinergic bronchodilator, which has hardly any anticholinergic side-effects when administered by inhalation [1–5]. In contrast to atropine and thiazinamium, OXBR and ipratropium bromide do not impair vagally induced mucociliary clearance, even at doses that produce maximal bronchodilatation [6]. Because the action of OXBR is expected to be more prolonged (up to 8 h) than that of ipratropium bromide, successful application in nocturnal asthma is anticipated [7, 8].

The only pharmacokinetic studies have been based on use of ^{14}C-labelled OXBR in the rats, dogs and humans. For anticholinergic drugs, such as oxyphenonium, ipratropium bromide, scopolamine and atropine, sensitive radioreceptor assays were developed for use in pharmacokinetic studies [9–12]. Although sensitivity was expected to be a problem in the analysis of plasma samples collected after oral administration and inhalation of OXBR, it was decided to develop a radioreceptor assay for the drug, and to attempt to improve its sensitivity. The aim of the present single dose study was to determine the bioavailability and pharmacokinetics of OXBR after different routes of administration. A comparison was also made between the plasma levels and urinary excretion after inhalation and oral administration, in order to assess the possible oral contribution to the anticholinergic effects observed after inhalation.

Materials and methods

Chemicals

^{3}H-NMS ^{3}H-N-methylscopolamine (85 Ci/mmol) was supplied by NEN (Dreieich, FRG). Oxitropium bromide was kindly donated by Boehringer Ingelheim KG (Ingelheim, FRG). Tetrapentylammonium iodide was obtained from Eastman Kodak (Rochester, NY, USA). All other chemicals and solvents of analytical grade were obtained from Merck (Amsterdam, The Netherlands). Polyethylene tubes were obtained from Greiner (Alphen a.d. Rijn, The Netherlands) and Sovirel tubes from Quickfit S.A. (Epernon, France). The 50 mM sodium phosphate buffer