Disposition and metabolism of codeine after single and chronic doses in one poor and seven extensive metabolisers

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- 1 The pharmacokinetics, metabolism and partial clearances of codeine to morphine, norcodeine and codeine-6-glucuronide after single (30 mg) and chronic (30 mg 8 h for seven doses) administration of codeine were studied in eight subjects (seven extensive and one poor metaboliser of dextromethorphan). Codeine, codeine-6-glucuronide, morphine and norcodeine were measured by high performance liquid chromatographic assays.
- 2 After the single dose, the time to achieve maximum plasma codeine concentrations was 0.97 ± 0.31 h (mean $\pm$ s.d.) and for codeine-6-glucuronide it was 1.28 ± 0.49 h. The plasma AUC of codeine-6-glucuronide was 15.8 ± 4.5 times higher than that of codeine. The AUC of codeine in saliva was 3.4 ± 1.1 times higher than that in plasma. The elimination half-life of codeine was 3.2 ± 0.3 h and that of codeine-6-glucuronide was 3.2 ± 0.9 h.
- 3 The renal clearance of codeine was 183 ± 59 ml min⁻¹ and was inversely correlated with urine pH ($r = 0.81$). These data suggest that codeine undergoes filtration at the glomerulus, tubular secretion and passive reabsorption. The renal clearance of codeine-6-glucuronide was 55 ± 21 ml min⁻¹, and was not correlated with urine pH. Its binding to human plasma was less than 10%. These data suggest that codeine-6-glucuronide undergoes filtration at the glomerulus and tubular reabsorption. This latter process is unlikely to be passive.
- 4 After chronic dosing, the pharmacokinetics of codeine and codeine-6-glucuronide were not significantly different from the single dose pharmacokinetics.
- 5 After the single dose, $86.1 \pm 11.4\%$ of the dose was recovered in urine, of which $59.8 \pm 10.3\%$ was codeine-6-glucuronide, $7.1 \pm 1.1\%$ was total morphine, $6.9 \pm 2.1\%$ was total norcodeine and $11.8 \pm 3.9\%$ was unchanged codeine. These recoveries were not significantly different ($P > 0.05$) after chronic administration.
- 6 After the single dose, the partial clearance to morphine was 137 ± 31 ml min⁻¹ in the seven extensive metabolisers and 8 ml min⁻¹ in the poor metaboliser; to norcodeine the values were 103 ± 33 ml min⁻¹ and 90 ml min⁻¹; to codeine-6-glucuronide the values were 914 ± 129 ml min⁻¹ and 971 ml min⁻¹; and intrinsic clearance was 1568 ± 103 ml min⁻¹ and 1450 ml min⁻¹. These values were not significantly ($P > 0.05$) altered by chronic administration. In the seven extensive metabolisers the values of partial clearance to morphine were not significantly ($P > 0.05$) different from those to norcodeine.
- 7 The usefulness of measuring partial clearances was illustrated by the detection of the poor oxidative metaboliser to morphine, whereas the measurement of parent compound in plasma did not identify this important metabolic defect.
- 8 Poor metabolisers of codeine may not derive any of the pharmacological effects associated with codeine.

Keywords codeine disposition codeine-6-glucuronide renal clearances morphine norcodeine partial clearances pharmacogenetics

Introduction

Codeine is a widely used analgesic, antitussive and anti-diarrhoeal drug, and has been available for more than 100 years (Jaffe & Martin, 1985). After administration to man, it is metabolised by *O*- and *N*-demethylation and glucuronidation (Adler, 1954; Adler *et al.*, 1955; Nomof *et al.*, 1977; Oberst, 1941; Yue *et al.*, 1989a,b) (Figure 1). The *O*-demethylation step is of particular importance, since it results in the formation of morphine, the currently accepted analgesic moiety (Jaffe & Martin, 1985). The pharmacokinetics of codeine in man have been studied by several investigators (Findlay *et al.*, 1977, 1978, 1986; Guay *et al.*, 1987, 1988; Quiding *et al.*, 1986; Rogers *et al.*, 1982). However, there are problems in the interpretation of the data derived from these studies. Firstly, all the studies except one (Quiding *et al.*, 1986) were based on the same radioimmunoassay (r.i.a.) (Findlay *et al.*, 1977) for the estimation of plasma codeine and morphine concentrations. Whilst the cross-reactivity of this assay for codeine metabolites was low, the high circulating codeine-6-glucuronide concentrations (*vide infra*) may well cause spuriously high plasma and urine codeine concentrations. Previous data for morphine, based on r.i.a., have been shown to be unreliable when compared with data obtained with specific high performance liquid chromatographic (h.p.l.c.) assays (Aherne & Littleton, 1985; Hanks & Aherne,

1985). Secondly, in all studies to date, codeine-6-glucuronide, a potentially important metabolite of codeine, by analogy with the pharmacologically active morphine-6-glucuronide (Osborne *et al.*, 1988; Pasternak *et al.*, 1987; Yoshimura *et al.*, 1973), was not determined directly. The concentrations of codeine-6-glucuronide in plasma were estimated by analysing codeine after β -glucuronidase hydrolysis (Findlay *et al.*, 1977, 1978, 1986; Guay *et al.*, 1987, 1988; Rogers *et al.*, 1982). It is known, however, that β -glucuronidase cannot completely hydrolyse codeine-6-glucuronide (Axelrod & Inscoe, 1960; Bodd *et al.*, 1987; Chen *et al.*, 1989b; Guay *et al.*, 1988; Yoshimura *et al.*, 1968). The concentrations of codeine-6-glucuronide in the previous studies may therefore have been under-estimated. Thirdly, single and chronic dose pharmacokinetics of codeine and the derived codeine-6-glucuronide have not been compared adequately. Quiding and coworkers (1986) compared the pharmacokinetics of codeine after single and chronic dosing of 60 mg, but little information could be obtained from the single dose pharmacokinetics because the sampling period was only 2.8 h. Whilst Guay and coworkers (1987) collected blood samples for 24 h, the r.i.a. used and hydrolysis by β -glucuronidase make the data from their study difficult to interpret. Nevertheless, they found no differences between the

Figure 1 Metabolic pathways of codeine.

areas under the plasma concentration-time curves of codeine and 'codeine glucuronide' after single and chronic administration, but found a significant increase in the area under the plasma concentration-time curve of morphine after chronic dosing. Fourthly, there is little information regarding the mechanism(s) of the renal clearance of codeine and codeine-6-glucuronide because most of the previous investigators collected blood samples only.

We and others (Chen *et al.*, 1988a; Dayer *et al.*, 1988; Yue *et al.*, 1989b) have shown that the *O*-demethylation of codeine to morphine is under genetic control and co-segregates with the debrisoquine/sparteine oxidative polymorphism. *In vivo* pharmacogenetic studies of codeine metabolism have relied on determining the metabolic ratio by measuring the metabolites and parent compound in urine. This approach provides only limited insight into a person's capacity to eliminate codeine through its various metabolic pathways. Partial clearance rates are considered to be better indices of *in vivo* drug metabolism (Jackson *et al.*, 1986).

The objectives of the present study were to determine the pharmacokinetics of codeine and codeine-6-glucuronide and the metabolism and partial clearances of codeine in human (one poor and seven extensive metabolisers) subjects given single and chronic oral doses of codeine and using sensitive and specific analytical methods.

Methods

Subjects

Eight healthy subjects, seven male and one female, participated in the study, which was approved by the Human Ethics Committee of the Royal Adelaide Hospital and the Committee on the Ethics of Human Experimentation of the University of Adelaide. The subjects were aged 27.3 ± 5.2 (mean $\pm$ s.d., range 25–37) years and weighed 67.9 ± 11.9 (range 48–83) kg. They were in good health as judged by physical and laboratory examinations. Four subjects smoked cigarettes. Other drugs and alcohol were prohibited for 2 days before codeine administration and for the duration of the study. The oxidative phenotype of these subjects was determined four weeks after the completion of the study, using dextromethorphan as the model drug (Chen *et al.*, 1990). Subject 2 was a poor metaboliser (metabolic ratio = 0.38) and the remainder were extensive metabolisers (metabolic ratio = 160–3460).

Study design

Single dose After an overnight fast, each subject received a single 30 mg codeine phosphate tablet (Fawns & McAllan, Croydon, Australia) followed by 100 ml of water. Food and fluid were permitted after 3 h. The subjects were ambulant but confined to the laboratory for the 12 h of sampling. Venous blood samples (10 ml) were collected through an indwelling catheter kept patent with a stylet (JelcoTM, Critikon Inc., Tampa, USA), placed in a forearm vein, into heparinized plastic

tubes at 0, 0.25, 0.5, 0.75, 1, 1.25, 1.5, 2, 3, 4, 6, 8 and 12 h after administration. At the same times, mixed saliva samples (3 ml) were collected by chewing ParafilmTM. All urine was collected in 0–12, 12–24, and 24–48 h aliquots after dosing.

Chronic dosing The second part of the study was conducted in the same subjects 2 to 3 weeks after completion of the single dose study. Subjects received 30 mg codeine phosphate as tablets from the same batch as above 8 hourly (08.00, 16.00, 24.00 h) for seven doses. The final dose was given at 08.00 h on day 3 after an overnight fast. Blood and saliva samples were collected at 0, 0.25, 0.5, 0.75, 1, 1.25, 1.5, 2, 4, 6 and 8 h after the last dose and urine was collected over the dosing interval (0–8 h).

Plasma was separated from blood cells by centrifugation. Urine pH and volume were recorded and an aliquot (10 ml) retained for analysis. Saliva pH was recorded. All samples were stored in stoppered vials at -20°C until analysis.

Drug analysis in biological fluids

Unconjugated codeine, norcodeine and morphine These were measured in urine, plasma (codeine only) and saliva (codeine only) by an h.p.l.c. method (Chen *et al.*, 1989a). The limit of assay was 2 ng ml^{-1} for morphine (7.0 nmol l^{-1}), norcodeine (7.0 nmol l^{-1}) and codeine (6.7 nmol l^{-1}) in plasma.

Codeine-6-glucuronide Codeine-6-glucuronide in plasma and urine was measured directly using an h.p.l.c. assay (Chen *et al.*, 1989b). The assay was also applied to saliva samples. The limit of assay was 10 ng ml^{-1} (21 nmol l^{-1}) for plasma and $0.1\text{ }\mu\text{g ml}^{-1}$ ($0.21\text{ }\mu\text{mol l}^{-1}$) for urine.

Morphine glucuronides Morphine-3- and -6-glucuronides in urine were measured directly using a modified h.p.l.c. method (Chen *et al.*, 1989b; Svensson *et al.*, 1982). The column ($150 \times 4.6\text{ mm i.d.}$) was packed with Spherisorb (5 μm ODS-2, Phase Separations, Queensferry, U.K.). The mobile phase comprised 26% (v/v) acetonitrile in 10 mM phosphate buffer (pH 2.1) containing 1 mM dodecyl sulphate. The flow rate through the column was 1 ml min^{-1} . The excitation and emission wavelengths of the fluorescence detector (LS-5 luminescence spectrometer, Perkin-Elmer, Beaconsfield, U.K.) were 230 and 350 nm, respectively. The urine sample was treated in the same manner as described for plasma codeine-6-glucuronide (Chen *et al.*, 1989b) except that the internal standard was normorphine, which itself could not be detected (less than $0.1\text{ }\mu\text{g ml}^{-1}$) in the urine of any of the subjects following both dosage regimens. The limit of assay for morphine-3-glucuronide and morphine-6-glucuronide was $0.1\text{ }\mu\text{g ml}^{-1}$ in urine.

Norcodeine conjugates The presence of conjugates of norcodeine in urine was inferred by the generation of norcodeine following enzymatic hydrolysis. The internal standard (dihydrocodeine) was added to urine samples (0.3 ml) in 10 ml screw capped tubes. β -glucuronidase from *Helix pomatia* type H-1 (Sigma Chemical Co., St

Louis, MO., USA) was added (0.5 ml of a 5000 u ml⁻¹ in pH 5.0 acetate buffer), followed by incubation in a shaking water bath at 37° C for 16 h. Samples were analysed for norcodeine by the method described above for unconjugated norcodeine (Chen *et al.*, 1989a).

Creatinine analysis

Creatinine concentrations in plasma and urine were measured by an h.p.l.c. method (Huang & Chiou, 1983). The coefficients of variation of all assays in terms of reproducibility and accuracy were less than 10%.

Protein binding of codeine-6-glucuronide

Drug-free plasma was spiked with codeine-6-glucuronide to achieve concentrations of 210, 525, 1050, 2100 and 10500 nmol l⁻¹ and mixed for 10 min at 37° C. Aliquots (1 ml) of the samples were then centrifuged using a Centrifree™ (Amicon Corporation, Danvers, MA, USA) ultrafiltration unit at 2000 g for 30 min at 37° C. Aqueous solutions of codeine-6-glucuronide in the same concentrations were treated in the same way. Aliquots of the ultrafiltrate were analysed for codeine-6-glucuronide by h.p.l.c. (Chen *et al.*, 1989b). The unbound fraction of codeine-6-glucuronide was calculated as the concentration of the ultrafiltrate in the plasma sample divided by the ultrafiltrate concentration in the aqueous sample (to exclude nonspecific binding).

Pharmacokinetic and statistical analyses

The concentrations of codeine in saliva up to 1 h after administration were not included in the calculations of the pharmacokinetic data, because of likely retention of part of the oral dose in the mouth during this time period.

The maximum plasma concentration ($C_{\max}$) and its time of occurrence ($t_{\max}$) were determined from the observed data. The elimination rate constant (λ_z) was calculated from the slope ($\times 2.303$) of the terminal portion of the semilogarithmic plasma concentration-time curve using linear regression analysis. The area under the plasma or saliva concentration-time curves to the last sampling time following the single dose (AUC

(0,12)) and the interdosing interval at steady state after multiple dosing (AUC_{ss}) was calculated by the linear trapezoidal rule and the total AUC for the single dose was calculated as:

$$AUC = AUC(0,12) + C(12)/\lambda_z^P \text{ or } C_{sal}(12)/\lambda_z^{sal}$$

where $C(12)$ and $C_{sal}(12)$ are the plasma and saliva concentrations at 12 h respectively. Half-lives ($t_{1/2,z}$) in plasma and saliva were calculated as:

$$t_{1/2,z} = 0.693/\lambda_z^P \text{ or } \lambda_z^{sal}$$

Renal clearance (CL_R) was calculated as:

$$CL_R = Ae(t_1,t_2) / AUC(t_1,t_2)$$

where $Ae(t_1,t_2)$ is the amount excreted unchanged over the urine collection interval (0–12 h single dose and 0–8 h chronic dosing).

Partial metabolic clearances (CL_m) of codeine to its measurable metabolites (morphine, norcodeine, codeine-6-glucuronide) were calculated as:

$$CL_m = Ae/AUC \text{ or } Ae/AUC_{ss}$$

where for morphine, Ae is the sum (in mols) of morphine, morphine-3-glucuronide and morphine-6-glucuronide excreted in urine; for norcodeine, Ae is the sum of norcodeine and norcodeine conjugates in urine and for codeine-6-glucuronide, Ae is the amount of this metabolite in urine. Intrinsic clearance was calculated as dose/AUC (single dose) or dose/ AUC_{ss} (chronic dosing).

Differences in pharmacokinetic data between treatments were analysed for statistical significance by the paired *t*-test. Statistical significance was assumed when $P < 0.05$. Linear regression analysis was also used. All data are tabulated as mean $\pm$ s.d.

Results

Codeine

The mean plasma and saliva codeine concentration-time profiles in eight subjects following single and chronic oral doses are shown in Figure 2. Tables 1 and 2 contain the derived pharmacokinetic parameters of codeine after single and chronic dosing, respectively. The mean $t_{\max}$ occurred in about 1 h after both single and chronic dosing ($P > 0.05$). The mean plasma $C_{\max}$ of codeine

Table 1 Pharmacokinetic parameters of codeine after a single 30 mg oral dose

Subject	$t_{1/2,z}$ (h)		$C_{\max}$ (nmol l ⁻¹)	$t_{\max}$ (h)	AUC (nmol l ⁻¹ h)		CL_R (ml min ⁻¹)	* f_e (%)
	plasma	saliva			plasma	saliva		
1	3.22	2.82	226	0.75	789	1926	178	11.1
2#	2.77	2.74	274	0.50	876	3665	157	10.8
3	3.63	3.11	138	1.00	816	2019	67	4.3
4	3.17	3.19	159	1.00	722	2601	167	9.5
5	2.81	3.18	186	1.25	856	2539	208	14.0
6	3.15	2.98	172	0.75	809	3062	260	16.6
7	3.60	2.72	142	1.50	886	2422	189	13.2
8	3.59	4.13	160	1.00	813	1726	238	15.2
Mean	3.24	3.11	182	0.97	821	2495	183	11.8
s.d.	0.34	0.45	46	0.31	53	638	59	3.9

* f_e : % of dose excreted unchanged in urine.

poor metaboliser.

Figure 2 Means $\pm$ s.d. plasma codeine ($\circ$) and codeine-6-glucuronide ($\bullet$) concentrations after a single 30 mg dose (a) and 30 mg 8 h for seven doses (b) of codeine phosphate.

after chronic dosing was significantly higher than that after single dosing ($P < 0.05$). The mean elimination half-lives determined from plasma and saliva after single and chronic dosing were not significantly different. There were no differences between the half-lives of codeine after the two treatments. The mean plasma codeine AUC for the single dose was not significantly different from that during the dosing interval at steady state. The mean saliva codeine AUC for the single dose was significantly lower than that following chronic dosing ($P < 0.05$). The saliva to plasma codeine AUC ratios after single and chronic dosing were 2.95 ± 0.72 and 3.75 ± 1.38 , respectively ($P < 0.05$). There was a positive correlation between saliva and plasma codeine concentrations ($r = 0.80$, $P < 0.01$; Figure 3) and an inverse correlation ($r = 0.83$, $P < 0.01$) between the saliva codeine AUC and saliva pH.

Codeine-6-glucuronide

The mean plasma codeine-6-glucuronide concentration-time profiles after single and chronic dosing are shown in Figure 2. Table 3 contains the derived pharmacokinetic parameters after both dosing regimens. There was no difference in the plasma $t_{\max}$ between codeine-6-glucuronide and codeine ($P > 0.05$) and for codeine-6-glucuronide there was no significant difference in this parameter between single and chronic dosing. The elimination half-lives of codeine-6-glucuronide averaged 3.2 ± 0.9 h after single dosing and 3.3 ± 0.6 h after chronic dosing and were not significantly different from that of codeine. The mean plasma AUC of codeine-6-glucuronide after the single dose was not significantly different from that during the dosing interval at steady state. The mean plasma AUC of codeine-6-glucuronide after the single

Table 2 Pharmacokinetic parameters of codeine after 30 mg 8 hourly orally for seven doses

Subject	$t_{1/2,z}$ (h)		$C_{\max}$ (nmol l ⁻¹)	$t_{\max}$ (h)	AUC_{ss} (nmol l ⁻¹ h)		CL_R (ml min ⁻¹)	* f_e (%)
	plasma	saliva			plasma	saliva		
1	3.13	3.81	239	0.75	792	1775	191	11.9
2#	3.37	3.29	291	1.25	1054	5919	201	16.7
3	3.25	2.84	243	0.75	786	2224	250	15.4
4	2.78	2.20	223	0.75	930	4739	219	16.0
5	2.63	2.35	228	1.00	786	3592	100	6.2
6	2.44	2.39	293	1.25	846	3926	246	16.3
7	2.91	2.48	265	0.75	1120	3288	265	23.4
8	2.68	2.17	265	0.75	913	1900	110	7.9
Mean	2.90	2.69	**256	0.91	903	**3421	198	14.2
s.d.	0.33	0.59	27	0.23	127	1451	63	5.5

* f_e : % of dose excreted unchanged in urine.

** significantly different ($P < 0.05$) from single dose (Table 1).

poor metaboliser.

Table 3 Pharmacokinetic parameters of codeine-6-glucuronide after single and chronic codeine dosing

Subject	$t_{1/2,z}$ (h)		C_{max} ($\mu\text{mol l}^{-1}$)		t_{max} (h)		$*AUC$ ($\mu\text{mol l}^{-1} \text{h}$)		CL_R (ml min^{-1})	
	S	C	S	C	S	C	S	C	S	C
1	2.95	4.43	3.23	2.57	1.25	1.00	12.98	12.27	57.1	74.7
2#	2.31	3.16	4.12	3.37	0.75	1.00	15.62	13.55	54.6	64.0
3	3.57	2.31	1.75	2.21	1.00	1.00	8.06	7.96	101.0	111.2
4	2.75	3.12	2.78	2.54	2.00	2.00	15.42	12.14	32.3	46.5
5	2.39	3.06	3.64	2.98	2.00	0.75	15.18	11.74	52.9	51.7
6	4.71	3.39	2.13	2.78	1.25	1.25	14.11	11.74	59.6	54.6
7	4.32	3.43	2.13	2.77	1.25	1.00	24.78	12.61	36.2	53.8
8	2.76	3.23	3.43	3.89	0.75	1.00	13.44	14.01	45.6	32.2
Mean	3.22	3.27	3.03	2.89	1.28	1.13	14.95	12.00	54.9	61.1
s.d.	0.89	0.58	0.78	0.53	0.49	0.38	4.66	1.83	21.0	23.7

S: single codeine dose; C: chronic codeine dosing.

*: $AUC = AUC(0, \infty)$ (single dose) or AUC_{ss} (chronic dosing).

poor metaboliser

and chronic dosing was 15.8 ± 4.5 times greater than that of codeine. The unbound fraction of codeine-6-glucuronide in plasma was 0.92 ± 0.06 ($n = 10$) at concentrations between 210 and 10500 nmol l^{-1} . Codeine-6-glucuronide was not detected in saliva.

Renal clearance

The renal clearances of codeine and codeine-6-glucuronide were not significantly different between single and chronic dosing ($P > 0.05$, Tables 1, 2 and 3). There was a statistically significant inverse relationship between urine pH and renal codeine clearance ($r = 0.81$, $P < 0.01$) but no such relationship occurred for codeine-6-glucuronide ($r = 0.08$, $P > 0.05$) (Figure 4). There was no significant ($P > 0.05$) correlation between renal clearance and urine flow rate for both codeine ($r = 0.35$) and codeine-6-glucuronide ($r = 0.03$). The creatinine clearance in the eight subjects ranged from 90 to 132 ml min^{-1} .

Figure 3 Correlation between plasma and saliva codeine concentrations ($r = 0.80$, $P < 0.01$).

Urinary excretion

The urinary recovery of codeine, norcodeine, morphine and their conjugates as a percentage of the administered dose in the eight subjects after single and chronic dosing is shown in Table 4. There were no statistically significant differences ($P > 0.05$) after the single dose and during chronic dosing at steady state.

Partial clearance

The partial metabolic clearance of codeine to each metabolite was not significantly different ($P > 0.05$) after single and chronic administration (Table 5). The partial clearance to morphine showed the widest inter-individual variation from 5 to 190 ml min^{-1} . In subject 2, who had the lowest urinary recoveries of morphine and morphine glucuronides (Table 4), the clearance of codeine to morphine was substantially lower than in the

Figure 4 Correlation between the renal clearances of codeine ($\circ$), codeine-6-glucuronide ($\blacktriangle$) and urine pH. For codeine $r = 0.81$ ($P < 0.01$) and for codeine-6-glucuronide $r = 0.08$ ($P > 0.05$).

Table 4 Urinary excretion of codeine and metabolites as % of dose after single and chronic oral codeine administration

Subject	Codeine		C-6-G		Morphine		M-3-G		M-6-G		Norcodeine		N-C		Total	
	S	C	S	C	S	C	S	C	S	C	S	C	S	C	S	C
1	11.1	11.9	58.4	72.1	0.3	0.6	5.7	2.9	1.8	0.9	3.7	1.9	7.2	1.7	89.9	92.0
3	4.3	15.4	64.0	69.8	0.7	0.7	4.3	5.1	0.7	0.6	1.5	3.7	6.9	2.2	82.9	97.5
4	9.5	16.0	40.4	44.5	0.9	0.4	5.4	8.5	1.2	0.4	0.9	3.2	4.5	1.3	63.3	74.3
5	14.0	6.2	63.0	47.8	1.0	0.2	6.7	3.7	1.9	0.1	5.4	2.4	1.1	3.4	93.4	63.8
6	16.6	16.3	66.3	50.5	0.4	0.6	6.6	9.8	1.2	1.4	4.7	4.5	2.4	4.2	99.1	87.3
7	13.2	23.4	70.5	53.4	1.0	0.9	4.4	6.9	1.0	0.8	3.8	4.4	1.8	2.5	95.8	92.3
8	15.2	7.9	48.4	35.6	1.3	1.0	7.4	10.7	2.1	1.9	2.8	2.1	1.8	1.1	79.6	60.3
Mean	12.0	13.9	58.7	53.4	0.8	0.6	5.8	6.8	1.4	0.9	3.3	3.2	3.7	2.3	86.3	81.1
s.d.	4.1	5.8	10.7	13.3	0.4	0.3	1.2	3.0	0.5	0.6	1.6	1.1	2.5	1.1	12.3	14.9
2*	10.8	16.7	67.0	68.3	0.0	0.0	0.6	0.4	0	0	1.9	9.3	4.6	1.0	84.9	95.5

C-6-G: codeine-6-glucuronide; M-3-G: morphine-3-glucuronide; M-6-G: morphine-6-glucuronide; N-C: norcodeine conjugates; S: single dose; C: chronic dosing.

*: poor metaboliser.

Table 5 Intrinsic clearance and partial clearances of codeine to its measurable metabolites after single and chronic codeine administration

Subject	Morphine		Partial clearance (ml min ⁻¹)				Intrinsic clearance (ml min ⁻¹)	
			Norcodeine		Codeine-6-glucuronide			
	single	chronic	single	chronic	single	chronic	single	chronic
1	154	70	166	55	940	1153	1610	1604
3	97	103	124	91	995	1128	1557	1616
4	141	128	91	58	709	606	1759	1366
5	148	65	91	88	946	773	1484	1616
6	145	177	105	123	1040	758	1570	1501
7	93	98	77	74	1010	607	1434	1134
8	180	190	68	42	756	495	1562	1391
Mean	137	119	103	76	914	789	1568	1461
s.d.	31	49	33	27	129	258	103	178
2*	8	5	90	117	971	825	1450	1205

*: poor metaboliser.

other subjects after both regimens (Table 5). The partial clearance to norcodeine was not significantly different than that to morphine ($n = 7$). The partial clearance to codeine-6-glucuronide showed the least degree of variation between subjects. There was no significant difference in the intrinsic clearance of codeine between single and chronic doses.

There were no adverse effects of codeine in any of the subjects during the studies.

Discussion

The values of the pharmacokinetic parameters for codeine after a single dose are similar to those reported previously (Findlay *et al.*, 1977, 1978, 1986; Quiding *et al.*, 1986; Rogers *et al.*, 1982). The plasma elimination half-life found by Guay and coworkers (1987) of 4.5–5.6 h is somewhat longer than those found by us and others. This difference may have resulted from the nonspecific r.i.a. used in Guay's study. The pharmacokinetics of codeine after multiple doses have been reported previously (Guay *et al.*, 1987; Quiding *et al.*, 1986). In contrast to the results of Guay and coworkers (1987), the

maximum plasma concentration of codeine in this study increased significantly after multiple dosing compared to the single dose and this is in agreement with the findings of Quiding and coworkers (1986). This can be explained by the presence of residual codeine in plasma from previous doses. Steady-state had been achieved since the zero and 8 h plasma codeine concentrations were not significantly different. Except for the increase in maximum concentration after chronic dosing, it would appear that the absorption and disposition of codeine are not altered by chronic administration, at least in the short term and at the doses studied.

The saliva to plasma codeine concentration ratio was greater than unity, confirming the report by Lee and coworkers (1986). Several properties of codeine would promote its penetration and retention in saliva. Codeine is a basic compound with a high pK_a (8.2) and low (7–25%) plasma protein binding (Moffat *et al.*, 1986). It is also highly lipophilic (octanol/water partition coefficient 3.98; Moffat *et al.*, 1986). The inverse correlation between saliva pH and saliva codeine AUC is in keeping with the theoretical prediction based on the Henderson-Hasselbalch equation. Pholcodine, a drug with similar physicochemical properties to codeine, also has a high

saliva to plasma concentration ratio (Chen *et al.*, 1988b). The finding that the saliva codeine AUC during the dosing interval at steady state was higher than that after the single dose may be due to the difference ($P = 0.05$) in saliva pH between the two treatments, which averaged 6.6 and 6.4 after the single and chronic dose, respectively. Codeine-6-glucuronide was not detected in saliva, a likely result of its high polarity and poor lipophilicity.

The renal clearance of codeine ranged from 67 to 265 ml min⁻¹. Since the degree of plasma binding is small (7–25%; Moffat *et al.*, 1986), and as the subjects' creatinine clearances ranged from 90 to 132 ml min⁻¹, these data indicate that in addition to glomerular filtration, codeine appears to undergo active secretion into the lumen of the proximal tubule. The significant negative correlation between renal clearance and urine pH suggests that passive reabsorption in the distal tubule/collecting duct also occurred as suggested by the observations of Vaughan & Beckett (1973).

We have confirmed the results of others (Adler *et al.*, 1955; Nomof *et al.*, 1977; Oberst, 1941; Yue *et al.*, 1989a,b) that codeine-6-glucuronide is the major metabolite of codeine in man. However, to our knowledge, ours is the first study to directly determine codeine-6-glucuronide disposition after codeine administration in humans. The pharmacokinetics of codeine glucuronide have only been studied indirectly by Guay and coworkers (1987, 1988) using β -glucuronidase hydrolysis and r.i.a. In their study, concentrations of the glucuronide were corrected to reflect the intrinsically less than complete efficiency of enzyme hydrolysis (Guay *et al.*, 1988). In addition, the efficiency of hydrolysis can be further reduced in biological media (Chen *et al.*, 1989b) because of enzyme inhibitors which are naturally present in biological fluids (Boyland & Williams, 1960; Combie *et al.*, 1982; Levvy, 1952). The mean apparent plasma half-life of codeine-6-glucuronide in our subjects was 3.2 h which was not significantly different from that of codeine, reflecting formation-dependent clearance of this conjugate. The renal clearance of codeine-6-glucuronide was much lower than creatinine clearance. Since the plasma binding of codeine-6-glucuronide was only 8%, the renal clearance by filtration would be between 83 and 121 ml min⁻¹. In seven of the eight subjects, the renal clearance of codeine-6-glucuronide was substantially lower than these values, suggesting that it was filtered and reabsorbed. However, other mechanisms need to be considered. For example, hydrolysis of codeine-6-glucuronide to codeine in the kidney and urinary tract could result in an inverse relationship between the apparent renal clearances of the two substances. No such relationship was found. Given the high polarity of the glucuronide, passive reabsorption is unlikely to occur and an active carrier-mediated process should be considered. We have demonstrated apparent reabsorption of morphine-3- and -6-glucuronide in the isolated perfused rat kidney (Sallustio *et al.*, 1989). The mechanism of this process for opioid glucuronides remains to be elucidated.

Our findings with respect to the urinary recovery of codeine and its major metabolites are in accord with those of Adler and coworkers (1955), Nomof and coworkers (1977), Vaughan & Beckett (1973) and Yue and coworkers (1989a,b).

The extent of *O*-demethylation of codeine showed extremely wide inter-individual variation considering the relative homogeneity of the group. Subject 2 is particularly noteworthy since only a small amount of total morphine (about 0.5% of dose) was recovered in urine after single and chronic administration of codeine. In this subject, the partial clearance to morphine was about one twentieth of the average of the other seven subjects. This subject was a poor metaboliser of dextromethorphan. This finding of a very low partial clearance to morphine, together with other *in vivo* (Chen *et al.*, 1988; Yue *et al.*, 1989b) and *in vitro* data (Dayer *et al.*, 1988; unpublished observations), confirms that the *O*-demethylation of codeine to morphine is under genetic control, cosegregating with the now well described debrisoquine/sparteine oxidative polymorphism (Eichelbaum *et al.*, 1979; Mahgoub *et al.*, 1977; Tucker *et al.*, 1977). Codeine can now be added to the list of those drugs whose oxidative metabolism is under pharmacogenetic control. Whilst this pathway for codeine is quantitatively minor, its potential importance cannot be underestimated since the analgesic effect of codeine is currently accepted as being mediated by the hepatic formation of morphine (Jaffe & Martin, 1985; Sanfilippo, 1948). Thus it would appear that some patients (about 5–10% of the Caucasian population) may not derive analgesia from codeine. This hypothesis is currently being investigated.

The detection of the poor oxidative metaboliser in our study came about only because urinary metabolites of codeine were assayed. The determination of the pharmacokinetics of codeine in plasma alone would have missed this important phenomenon as this poor metaboliser had plasma codeine concentration-time profiles which were no different to those of the other seven subjects. This study highlights the need to characterize the metabolism of a drug in addition to its disposition in plasma.

Codeine is a drug of intermediate extraction (Rowland & Tozer, 1989) and this is the result primarily of conjugation at the -6- position. The demethylation clearance rates to morphine and norcodeine were similar in magnitude, and substantially lower (about 1/4) than for 6-glucuronidation. The partial clearances did not change after short term chronic administration, the way in which codeine is often administered clinically. At present, little is known about the factors which alter the metabolism of codeine in humans. Since the partial clearance to norcodeine was normal in the poor metaboliser, it would be reasonable to infer that the control of the two demethylation processes (*N*-demethylation to norcodeine, *O*-demethylation to morphine) is mediated by different cytochrome P-450 isoenzymes. It is important to identify the factors which regulate the demethylations and glucuronidation of codeine. With regard to *O*-demethylation of codeine, which is catalysed by cytochrome P-450 IID6 (Nebert *et al.*, 1989), genetic factors and drugs such as quinidine (a specific inhibitor of cytochrome P-450 IID6 (Brinn *et al.*, 1986)) are important. We are not aware of any information regarding factors which may alter the *N*-demethylation process. Several drugs have been shown to inhibit the glucuronidation of codeine in human liver microsomes (Yue *et al.*, 1990). Any factor which might alter the glucu-

ronidation of codeine and thus alter the proportion of the clearance of codeine to morphine could have important clinical implications.

Glucuronidation is generally regarded as a detoxification mechanism for xenobiotics. However, morphine-6-glucuronide has been documented to be a pharmacologically active compound and its analgesic potency is equal to or higher than that of morphine (Osborne *et al.*, 1988; Pasternak *et al.*, 1987; Yoshimura *et al.*, 1973). Whether codeine-6-glucuronide has pharmacological activity is unknown. The binding affinity of codeine-6-glucuronide to the μ -opioid receptor in rat brain homogenate is 300 times less than that of morphine and morphine-6-glucuronide, but similar to that of codeine (Irvine *et al.*, 1989). Plasma concentrations of codeine-6-glucuronide were 15 times higher than codeine and could be sufficient to contribute to analgesia and/or toxicity.

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