Pharmacokinetics of Prilocaine after Intravenous Administration in Volunteers

Enantioselectivity

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Background: Prilocaine exists in two stereoisomeric configurations, the enantiomers S(+)- and R(-)-prilocaine. The drug is clinically used as the racemate. This study examined the pharmacokinetics of the enantiomers after intravenous administration of the racemate.

Methods: Ten healthy male volunteers received 200 mg racemic prilocaine as a 10-min intravenous infusion. Blood samples were collected for 8 h after the start of the infusion. Plasma concentrations were measured by stereoselective high-performance liquid chromatography (HPLC). Unbound fractions of the enantiomers in blank blood samples, spiked with racemic prilocaine, were determined using equilibrium dialysis.

Results: The unbound fraction of R(-)-prilocaine (mean ± SD, 70% ± 8%) was smaller ($P < 0.05$) than that of S(+)-prilocaine (73% ± 5%). The total plasma clearance of R(-)-prilocaine (2.57 ± 0.46 l/min) was larger ($P < 0.0001$) than that of S(+)-prilocaine (1.91 ± 0.30 l/min). The steady-state volume of distribution of R(-)-prilocaine (279 ± 94 l) did not differ from that of S(+)-prilocaine (291 ± 93 l). The terminal half-life of R(-)-prilocaine (87 ± 27 min) was shorter ($P < 0.05$) than that of S(+)-prilocaine (124 ± 64 min), as was the mean residence time of R(-)-prilocaine (108 ± 30 min) compared with S(+)-prilocaine (155 ± 59 min; $P < 0.005$).

Conclusions: The pharmacokinetics of prilocaine are enantioselective. The difference in clearance is most likely a result of a difference in intrinsic metabolic clearance. The difference in the pharmacokinetics of the enantiomers of prilocaine does not seem to be clinically relevant. (Key words: Chiral drugs; protein binding; toxicity.)

THE amide-type local anesthetic prilocaine contains a chiral center and thus exists in two stereoisomeric configurations, the enantiomers S(+)- and R(-)-prilocaine. Except for lidocaine, which does not exhibit stereoisomerism, and ropivacaine, which is marketed as a single enantiomer, the local anesthetics of the amide type are clinically used as the racemate, containing equal amounts of both enantiomers. In general, enantiomers of drugs may differ in their pharmacologic properties. Studies of the differences in local anesthetic activity and toxicity of the enantiomers of several amide-type local anesthetics have first been published in the late 1960s and early 1970s. The topic regained attention in the early 1990s when publications emphasizing pharmacologic differences between the enantiomers of bupivacaine reappeared in the literature. Differences in the pharmacokinetics of the enantiomers after intravenous administration have been reported for bupivacaine and mepivacaine, but not for prilocaine.[1,2] The aim of this study was to get a detailed assessment of the enantioselectivity of the pharmacokinetics of prilocaine in humans.

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Materials and Methods

Subjects

The study protocol was approved by the Committee on Medical Ethics of the Leiden University Medical Center. Ten non-smoking, healthy male volunteers, aged 21–27 yr, body weight 69–85 kg, participated in the study after giving informed consent. Their state of health was confirmed by taking their medical history, physical examination, electrocardiogram, and laboratory tests,

which included urine analysis, hematology, and serum biochemistry. None of the volunteers ware taking medical or non-medical drugs. They refrained from food and drink (except water) from midnight before the experiment until 3 h after drug administration, when they received lunch.

Study Design

The study design was similar to that previously used in studies examining the enantioselectivity in the pharmacokinetics of bupivacaine and mepivacaine.[1,2] The experiments started between 8 and 10 AM and were performed in one of the operating rooms at the Leiden University Medical Center. Two antecubital intravenous catheters were introduced bilaterally, one for drug infusion and one for blood sampling. The volunteers, positioned supine, were monitored with continuous electrocardiogram and noninvasive blood pressure measurements until 1 h after drug infusion. After the collection of a blank 30-ml blood sample, 200 mg racemic prilocaine, diluted in 50 ml saline, was infused at a constant rate in 10 min. Blood samples for the determination of the plasma concentrations of the enantiomers were collected in heparinized glass tubes at 2, 5, 10, 15, 20, and 30 min after the start of the infusion, then at 15-min intervals until 120 min, at 30-min intervals until 240 min, and from then on at 1-h intervals until 8 h after the start of the infusion.

Laboratory Methods

Unbound fractions of the enantiomers of prilocaine were determined in duplicate in blank plasma samples, spiked with 1.5 μg/ml racemic prilocaine (0.75 μg/ml of each enantiomer) using equilibrium dialysis at 37°C, as described previously for bupivacaine, but using equal (1 ml) volumes of plasma and buffer solution.[3] Plasma concentrations of the enantiomers of prilocaine were measured using a stereoselective high-performance liquid chromatographic (HPLC) method, modified from Tucker et al.[4] Plasma (1 ml), to which ropivacaine (832 ng/ml) had been added as internal standard, was made alkaline by adding 50 μg 2 N NaOH and extracted with 5 ml di-ethylether. After centrifugation at 4°C the organic phase was extracted with 0.5 ml 0.1 N HCl. The aqueous phase was then washed with 5 ml n-pentane, made alkaline with 100 μl 2 N NaOH, and extracted with 5 ml n-pentane. After centrifugation at 4°C the organic phase was collected, evaporated to dryness in a stream of dry nitrogen at 40°C, and the residue dissolved in 100 μl of the mobile phase. The same procedure was followed to determine the concentrations in dialysate (1 ml, to

which 100 μl isoamylalcohol had been added). The HPLC system was equipped with a 15-cm (inner diameter, 0.46 cm) column, containing cellulose tris-3,5-dimethylphenyl carbamate, coated on silicagel, 5 μm (Chiralcel OD-H, J.T. Baker, Deventer, The Netherlands). The column temperature was 30°C. The mobile phase consisted of n-hexane, ethanol, and diethylamine (19:1:0.1), and the flow rate was 0.7 ml/min. Ultraviolet detection was at 238 nm. The injection volume was 30 μl. Calibration curves were constructed from blank blood samples, that were spiked with racemic prilocaine and were linear (r > 0.999) in the concentration range (20–2,000 ng/ml) covered. The retention times of ropivacaine, S(+)-prilocaine, and R(-)-prilocaine were 5.81 min, 7.11 min, and 7.82 min, respectively. The detection limit was approximately 5 ng/ml for each enantiomer, and the coefficient of variation did not exceed 10% in the concentration range encountered in this study.

Pharmacokinetic Analysis

The areas under the plasma concentration–time curves (AUC) and the first moment of these curves (AUMC) were determined using the linear trapezoidal rule when concentrations were increasing and using the logarithmic trapezoidal rule when concentrations were decreasing, with addition of the areas from the last sampling point until infinity.[5] Terminal rate constants (M_Z) were determined by linear regression analysis of the terminal part of the logarithm of the plasma concentration *versus* time curves from 120, 150, 180, 210, or 240 min on, including a minimum of four data points, covering a period of at least 3 h. Terminal half-lives ($t_{1/2,z}$), mean residence times (MRT), total plasma clearances (Cl), and volumes of distribution at steady-state (V_{ss}) were calculated as: $t_{1/2,z} = \ln(2) / M_Z$; MRT = AUMC / AUC - T / 2; Cl = dose / AUC and $V_{ss} = Cl \bullet MRT$, where T is the duration of the infusion.[5-7]

Statistical Analysis

Data were summarized as mean ± SD. Pharmacokinetic data of S(+)-prilocaine and R(-)-prilocaine were compared using the paired *t* test. P < 0.05 was considered statistically significant.

Results

The intravenous administration of racemic prilocaine was well tolerated by all volunteers. No signs or symptoms of systemic toxicity were observed.
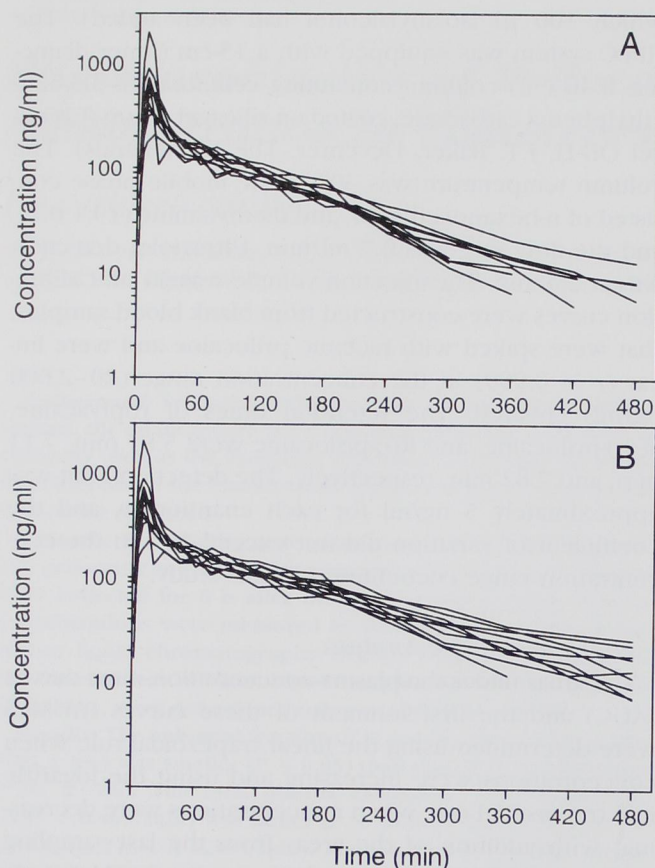
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Fig. 1. Plasma concentrations of R(-)-prilocaine (*A*) and S(+)-prilocaine (*B*) in individual subjects.

Total plasma concentrations of the individual enantiomers in all subject are shown in figure 1. Figure 2 shows the S(+):R(-) concentration ratio *versus* time for all subjects. The highest plasma concentrations were measured in the samples taken at the end of the infusion, except in three subjects, in whom the highest plasma concentrations were measured in the samples taken 5 min after termination of the infusion. No significant differences were found in the magnitude of the highest plasma concentrations of R(-)-prilocaine (mean ± SD, 670 ± 502 ng/ml) and S(+)-prilocaine (685 ± 516 ng/ml). The similarity of the concentrations of the enantiomers during the infusion (S(+)-:R(-)-prilocaine concentration ratios ranged from 0.91–1.08) indicates that the initial distribution characteristics of the enantiomers are similar. After termination of the infusion, plasma concentrations of S(+)-prilocaine and R(-)-prilocaine diverged in all subjects. The pharmacokinetic data in each subject and the mean ± SD of these parameters are presented in the table 1. Unbound fractions of S(+)-prilocaine were

slightly higher than those of R(-)-prilocaine, except in three subjects, in whom the unbound fractions of the enantiomers were equal. The volumes of distribution of the enantiomers did not differ significantly. S(+)-prilocaine was found to have a lower plasma clearance and a longer terminal half-life and mean residence time than R(-)-prilocaine.

Discussion

Laboratory studies on the qualities of the enantiomers of prilocaine showed that S(+)-prilocaine and R(-)-prilocaine have similar blocking effects on nerve conduction in isolated nerves.[8] However, when studied *in vivo* in guinea pigs, rats, and rabbits, S(+)-prilocaine exhibited a more pronounced local anesthetic effect and a longer duration of action compared with R(-)-prilocaine.[8] The authors suggested that this may be related to better, stereospecific, binding properties or different effects on the peripheral vascular bed, resulting in slower systemic absorption of S(+)-prilocaine. In toxicity studies no difference was found in the LD$_{50}$s of S(+)-prilocaine and R(-)-prilocaine after intravenous and subcutaneous administration of individual enantiomers in mice, although with continuous infusion in mice and after intermittent slow intravenous injections in rabbits, the toxic effects of S(+)-prilocaine were more pronounced than those of R(-)-prilocaine.[9] After intravenous injection of single enantiomers in cats, the maximal level of methemoglobinemia did not differ, although the rate of methemoglobin formation was more rapid after intravenous injection of R(-)-prilocaine compared with S(+)-prilocaine.[9] In cats
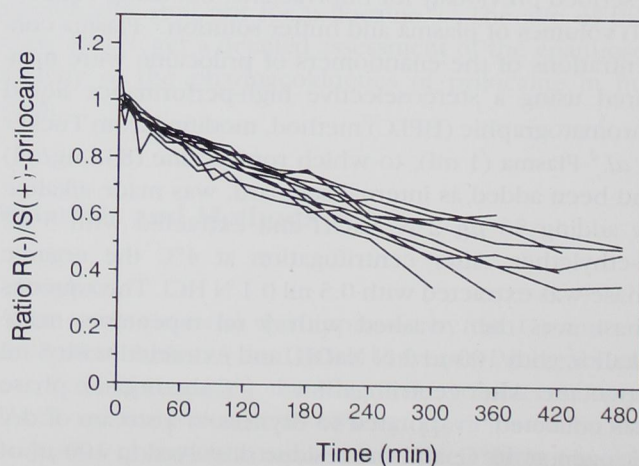

Fig. 2. Ratio of the plasma concentrations of R(-)-prilocaine and S(+)-prilocaine in individual subjects.

Table 1. Pharmacokinetic Data of R(−)- and S(+)-Prilocaine

Subject Number	f_u(R) (%)	f_u(S) (%)	Cl(R) (L/min)	Cl(S) (L/min)	V_{ss}(R) (L)	V_{ss}(S) (L)	$t_{1/2,z}$(R) (min)	$t_{1/2,z}$(S) (min)	MRT(R) (min)	MRT(S) (min)
1	75	76	2.77	2.29	253	259	74	88	91	113
2	58	65	2.35	1.51	297	419	133	293	126	278
3	82	82	3.41	2.39	348	324	55	80	102	136
4	69	69	2.40	1.89	266	271	95	117	111	143
5	68	76	1.82	1.55	112	126	70	88	62	81
6	73	74	2.68	1.91	471	446	124	162	176	233
7	72	73	2.49	1.77	311	305	92	117	125	172
8	61	70	2.05	1.65	197	215	101	119	96	130
9	78	78	2.65	2.02	243	247	68	83	92	122
10	60	68	3.06	2.10	290	296	56	91	95	141
Mean	70	73	2.57	1.91	279	291	87	124	108	155
SD	8	5	0.46	0.30	94	93	27	64	30	59
P	<0.05		<0.0001		NS		<0.05		<0.005	

f_u = unbound fraction in plasma; Cl = total plasma clearance; V_{SS} = steady-state volume of distribution; $t_{1/2,z}$ = terminal half-life; MRT = mean residence time; SD = standard deviation.

the plasma concentrations of S(+)-prilocaine were higher than those of R(-)-prilocaine on intravenous infusion of each enantiomer.[9] Hydrolysis rates of ^{14}C-labeled enantiomers of prilocaine by liver homogenates and slices of mice, rabbits, and cats and by microsomes of rabbits differed, indicating that R(-)-prilocaine had a higher affinity for the enzyme(s).[9] The authors concluded that no evidence was obtained that would suggest substitution of the racemate by either of its enantiomers to be advantageous. However, enantioselective disposition in humans can be an argument for substitution of the racemate by a single enantiomer. Tucker et al. reported slightly higher plasma concentrations of the S(+)-enantiomer compared with the R(-)-enantiomer after brachial plexus block with the racemate in six patients.[4] After oral intake of the racemate by four healthy volunteers, plasma concentrations of S(+)-prilocaine were found to be much higher compared to R(-)-prilocaine. The latter observation indicates a large difference in intrinsic metabolic clearance of the enantiomers through gut and liver.[4]

In our study, the plasma concentrations of each enantiomer during the infusion were similar in all volunteers, and no significant differences were present in the calculated volumes of distribution. Differences were found, however, in plasma clearance, and consequently in the terminal half-life and mean residence time, indicating a faster elimination of R(-)-prilocaine. The difference in clearance between the enantiomers can be explained by differences in enzyme affinity, which is in accordance with the studies of Åkerman and Ross[9] and Tucker et al.[4]

Pharmacokinetic data of prilocaine after intravenous administration in volunteers have been reported previ-

ously by Arthur et al.[10] Although they did not measure the plasma concentrations of the individual enantiomers but rather mixed enantiomers of prilocaine, their published pharmacokinetic data coincide with our findings. Of particular interest are the large plasma clearance values. In our study and in the study of Arthur et al., the plasma clearance of both enantiomers exceeded normal hepatic blood flow (1,200–1,700 ml/min),[11] suggesting some degree of extrahepatic clearance. A possible site of metabolism is the lung. Studying pulmonary retention of local anesthetics in humans, Kietzmann et al. found a concentration difference of prilocaine between mixed venous and arterial blood up to 15 min after epidural administration.[12] As retention was not anymore observed after 15 min, metabolic elimination of prilocaine by the lung in humans is unlikely.

Differences in potency, toxicity, and pharmacokinetics have been studied most extensively for the enantiomers of bupivacaine. The S(-)-enantiomer of bupivacaine appears to be favorable for clinical use because of its lower intrinsic toxicity,[13-18] and recently, a similar clinical efficacy has been demonstrated for S(-)-bupivacaine compared with racemic bupivacaine in epidural anesthesia.[19] A similar advantage in the clinical use of a single enantiomer of mepivacaine is less obvious.[13,20,21] The pharmacokinetic parameters of the enantiomers of bupivacaine and mepivacaine have been assessed in studies as described here for prilocaine.[1,2] The clearance and the volume of distribution at steady-state of the individual enantiomers of these local anesthetics differed. Clearance and volume of distribution at steady-state were respectively 25% and 56% larger for R(+)-bupivacaine compared with S(-)-bupivacaine and respectively 125%

and 79% larger for R(-)-mepivacaine compared with S(+)-mepivacaine. The differences in clearance and distribution could largely (with bupivacaine) or partly (with mepivacaine) be explained by differences in protein binding. Free fractions were 50% larger for R(+)-bupivacaine compared with S(-)-bupivacaine and 43% larger for R(-)-mepivacaine compared with S(+)-mepivacaine. This study also showed a significant difference in the degree of protein binding of the enantiomers of prilocaine too, but the difference was small and cannot explain the difference in the clearance of the enantiomers.

In conclusion, this study demonstrated enantioselectivity of the pharmacokinetics of prilocaine. The differences in clearance between the enantiomers of prilocaine seem to be a result of differences in enzyme affinity. On repeated injections or during continuous infusions the differences in the concentrations of the enantiomers could become substantial. However, because prilocaine is generally used for single injections because of the risk of induction of methemoglobinemia, the clinical relevance of the enantioselectivity in the pharmacokinetics of prilocaine is likely to be small.

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