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Spinal biopharmaceutics of bupivacaine and lidocaine by microdialysis after their simultaneous administration in rabbits

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Abstract

The aim of the present study was to determine the intrathecal bioavailability of a mixture of lidocaine and bupivacaine in a rabbit model of spinal anesthesia by using the microdialysis technique. Catheter and microdialysis probe were inserted under control of the view either in the epidural or in the intrathecal space. First, the epidural disposition of the mixture of bupivacaine and lidocaine was studied after epidural administration. Then, the intrathecal and plasma dispositions of bupivacaine and lidocaine were investigated following intrathecal or epidural administration. The epidural clearance of bupivacaine was higher than that of lidocaine, suggesting a more significant uptake of bupivacaine into the systemic circulation and/or into the CSF. The intrathecal bioavailability of bupivacaine and lidocaine was 12.3 and 17.9%, respectively, while it was 5.5 and 17.7% following the separate administration of each agent [Clément, R., Malinovsky, J.M., Le Corre, P., Dollo, G., Chevanne, F., Le Verge, R., 1999. Cerebrospinal fluid bioavailability and pharmacokinetics of bupivacaine and lidocaine following intrathecal and epidural administrations in rabbits using microdialysis. J. Pharmacol. Exp. Ther. 289, 1015–21]. After intrathecal administration, a decrease in C_{max} and AUC values was observed for bupivacaine in comparison with the separate administration. Moreover, after epidural administration, the systemic resorption was slower and lower, especially for bupivacaine. Such a reduction in the systemic absorption of bupivacaine might increase its intrathecal bioavailability, resulting from a vasoconstrictor effect of lidocaine reducing the systemic absorption of bupivacaine from the epidural space leading to an increase of its extent of absorption through meninges into CSF although its absorption rate was not modified.

Keywords: Microdialysis; Lidocaine–bupivacaine; Intrathecal administration; Epidural administration; Intrathecal bioavailability

1. Introduction

Mixtures of local anesthetics with short onset and long duration of action have been used to

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provide rapid and long lasting regional anesthesia. However, the duration of anesthesia of such mixture is different than those of each agent administered alone. Epidural (Cohen and Thrulow, 1979) or intradermal (Kim et al., 1970) bupivacaine provides a longer anesthesia than that of a mixture of choroprocaine and bupivacaine. An intermediate duration of anesthetic effects has been reported with a mixture of lidocaine and bupivacaine compared to bupivacaine administered alone in the epidural space (Kaulimen et al., 1980, Seow et al., 1982). In contrast, a clinical advantage in speed of onset, without significant shortening of duration of action, has been reported for a mixture of lidocaine and bupivacaine compared to bupivacaine alone after epidural administration (Magee et al., 1983).

The technique of in vivo microdialysis has found important applications in the field of pharmacokinetics, especially concerning the distribution and the metabolism of drugs (Elmquist and Sawchuk, 1997, Hansen et al., 1999). Recently, using the microdialysis technique, we reported differences in disposition of lidocaine and bupivacaine after their separate epidural injection in rabbits (Clément et al., 1999). The intrathecal bioavailability and the systemic uptake of lidocaine and bupivacaine following their separate epidural administration of an equimolar dose were significantly different.

Following epidural injection, local anesthetics cross the meninges to block spinal nerves and to induce anesthesia (Bromage et al., 1963). As duration of local anesthetics effects are related to the dose available to block conduction in spinal nerves, different durations of effects may be related to different intrathecal bioavailabilities. We speculated that the administration of a mixture containing different local anesthetics may modify the intrathecal bioavailability of each component following an epidural administration. Hence, the aim of the present study was to determine the intrathecal bioavailability of both lidocaine and bupivacaine in a rabbit model of spinal anesthesia following their simultaneous administration.

2. Materials and methods

2.1. Chemicals

Local anesthetics (bupivacaine, etidocaine, lidocaine and ropivacaine) were supplied by Astra (Astra Pain Control, Sweden). A Ringer's solution (NaCl 147 mM, KCl 4.4 mM, $CaCl_2$, $2H_2O$ 2.0 mM) was used as perfusion fluid. All other reagents were of analytical grade.

2.2. Animals

The study was approved by the local Committee of Laboratory Investigation and Animal Care and performed in accordance to French Ministry of Agriculture laws and guidelines for laboratory animal experiments. Experiments were performed on female New-Zealand albino rabbits, weighing 3.0 ± 0.3 kg, housed individually in standard cages with free access to food and water. Animals were fasted the night before experiment.

Throughout the experiment, animals were sedated with intermittent intravenous 1% thiopental. Insertion of spinal and vascular catheters were performed after L5–L6 laminectomy under epidural procaine anesthesia (Malinovsky et al., 1997).

After surgical incision of the meninges, the catheters and probes were inserted either in epidural or in intrathecal space, as described in Fig. 1. When catheter and microdialysis probe were not placed in the same space, they were inserted in order to put the tip of catheter opposite to the tip of the probe. A catheter was inserted via the femoral artery for blood sampling.

2.3. Microdialysis conditions and calibration

Microdialysis sampling was performed using a CMA/102 microinjection pump coupled to a microdialysis probe CMA/120 (membrane length 10 mm, 0.5 mm outer diameter, molecular weight cut off 20 kDa, CMA Microdialysis, Sweden). Microdialysis samples were collected during a 1 min interval every 2 min. Dialysates (sample volume = 1

μl) were collected in vials containing 100 μl of etidocaine (1 μg/ml) (external standard of HPLC) and a 50 μl aliquot was injected onto the chromatograph.

Retrodialysis, using ropivacaine as internal standard, was applied to calibrate the microdialysis probes. As previously shown, ropivacaine can be used as internal standard to study the disposition of bupivacaine (Clément et al., 1998) and of lidocaine (Clément et al., 1999). Thus, the probe was perfused throughout the experiment, at a flow rate of 1 μl/min with a Ringer's solution containing ropivacaine (100 μg/ml). During the experiment, The relative loss (RL) of ropivacaine was determined in each sample and used to correct the dialysate concentrations.

2.4. Chromatographic analysis

The separation and quantification of the local anesthetics in the dialysate (CSF, epidural samples) or in plasma samples were carried out using a high pressure liquid chromatography method with UV absorbance detection ($\lambda = 205$ nm). Dialysate samples were immediately injected onto the chromatographic system. The blood samples were centrifuged and plasma was stored frozen until analysis. Local anesthetics in plasma were extracted from plasma before analysis by HPLC according to a previously published method (Le Guévello et al., 1993). The limit of quantification of bupivacaine and lidocaine were 3 and 1.5 μg/ml in dialysate, and 4 and 2 ng/ml in plasma, respectively.

The chromatographic system consisted of a Waters Model 6000A pump (Waters Assoc., Milford, MA) equipped with a Waters Model 717 automatic injector, an LDC Milton Roy Model Spectromonitor 3100 variable-wavelength detector (LDC Milton Roy, Riviera Beach, FL), and a Delsi Model Enica 21 integrator (Delsi, Suresne, France). The analytical chromatographic column was a Lichrospher RP-B Merck (length 125 mm, internal diameter 3 mm). The flow rate was 0.5 ml/min, and the temperature was maintained at 30°C. The mobile phase consisted of a mixture of acetonitrile and pH 2.1, 0.01 M sodium dihydrogenphosphate.

2.5. Study design

In a first part, we investigated in three animals the epidural disposition of bupivacaine and lidocaine following their simultaneous epidural administration. Equimolar doses (6.9 μM, 2 mg of bupivacaine and 1.62 mg of lidocaine) were administered in 30 s under a volume of 1 ml. Two administrations separated by 2 h were performed leading to a serie of six concentration–time profiles for each drug. Epidural sampling was achieved every 2 min over 55 min.

In a second part, we investigated in three animals the intrathecal disposition of bupivacaine and lidocaine following their simultaneous intrathecal administration. Equimolar doses (0.2 μM, 0.060 mg of bupivacaine and 0.049 mg of lidocaine) were administered in 30 s under a vol-

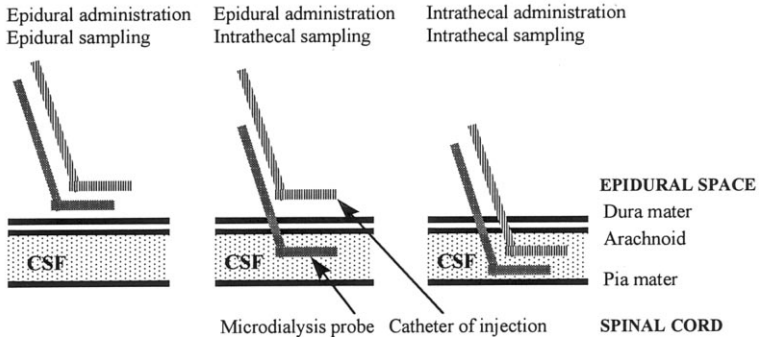

Fig. 1. Schematic representation of the insertion of the microdialysis probe and of the catheter of injection.

ume of 0.1 ml. Two administrations separated by 2 h were performed leading to a serie of six concentration–time profiles for each drug. Intrathecal sampling was achieved every 2 min over the 30 first min and then every 4 min over the following 24 min. Blood sampling (1.5 ml) was achieved in each animal according to the following schedule: before administration and then at 0.5, 1, 3, 5, 7, 11 and 15 min.

In a third part, we investigated in five animals the intrathecal and plasma dispositions of bupivacaine and lidocaine following their simultaneous epidural administration. Equimolar doses (6.9 μM, 2 mg of bupivacaine and 1.62 mg of lidocaine) were administered in 30 s under a volume of 1 ml. Two administrations separated by 2 h were performed leading to a serie of eight intrathecal concentration–time profiles for each drug. Five plasma concentration–time profiles were obtained following the first administration. Intrathecal sampling was achieved every 2 min over the first 30 min and then every 4 min over the following 24 min. Blood sampling (1.5 ml) was achieved in each animal according to the following schedule: before administration and then at 0.5, 1, 3, 5, 7, 9, 21, 29, 45 and 55 min.

2.6. Biopharmaceutic analysis

The maximum total plasma concentration and the maximum free epidural and intrathecal concentration (C_{max}) and the corresponding time (T_{max}) were derived from raw data. Areas under CSF or epidural concentration–time curves from the time of drug administration up to the last sampling point were computed by the linear trapezoidal rule by using a noncompartmental model with the software package WinNonlin (version 1.5, Scientific Consulting Inc., Apex, NC). Aera under epidural concentration–time curves was defined as AUC-epi. Aeras under CSF concentration–time curves after epidural and intrathecal administrations were defined as AUC-csf-epi and AUC-csf-it, respectively. Because both epidural and intrathecal administrations were not performed in the same animals, a mean CSF bioavailability (F-csf) was determined following:

$$\text{Fcsf} = (\text{mean AUC-csf-epi/epidural dose})\ /(\text{mean AUC-csf-it/intrathecal dose})$$

Individual CSF absorption kinetics constant (K_a) after epidural administration was determined by using the statistical pharmacokinetic software P-Pharm (version 1.5, Innaphase, Champs sur Marne, France).

The elimination clearance (CL) from the epidural space after epidural administration and from the intrathecal space after intrathecal administration was defined as the ratio between the dose administered and the aera under the concentration–time curves.

2.7. Statistics

Data are presented as mean ± S.D. and individual means were compared by using the Student's t test with a P value less than 0.05 considered as statistically significant.

3. Results and discussion

3.1. Epidural administration and epidural microdialysis

Individual epidural concentration–time profiles of bupivacaine and lidocaine following epidural administration of an equimolar bupivacaine–lidocaine mixture showed a biphasic decline (Fig. 2). The biopharmaceutic parameters are presented in Table 1. The C_{max} values were more variable but close to those obtained after separate administration of bupivacaine and lidocaine (2.45 ± 1.00 and 4.15 ± 1.18 mM, respectively) (Clément et al., 1999). The higher epidural clearance (CL) of bupivacaine compared to lidocaine suggested a more signifiant uptake of bupivacaine into the systemic circulation and/or into the CSF. Such a difference was also observed after separate epidural administration, although the clearance values were three times smaller.

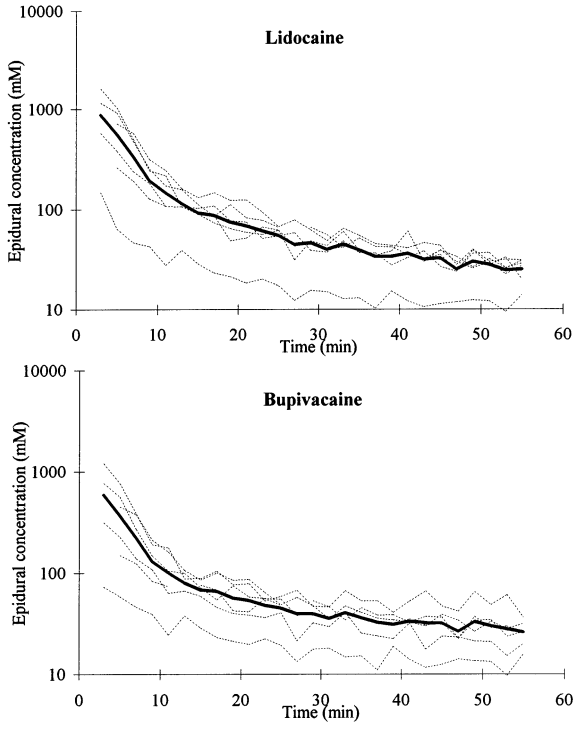

Fig. 2. Individual and mean epidural concentrations of bupivacaine (bottom) and lidocaine (top) after epidural administration of an equimolar bupivacaine–lidocaine mixture (6.9 μM) in rabbits.

Table 1
Epidural biopharmaceutic parameters (mean ± S.D.) of bupivacaine and lidocaine after epidural administration of an equimolar bupivacaine–lidocaine mixture (6.9 μM) in rabbits

		Bupivacaine	Lidocaine
C_{max}	(μg/ml)	491 ± 422	745 ± 557**
	(mM)	1.70 ± 1.47	3.15 ± 2.36
AUC-epi	(mM min)	20.9 ± 13.4	36.6 ± 19.9**
CL	(ml/min)	0.525 ± 0.450	0.281 ± 0.237*

* $P<0.05$.
** $P<0.01$.

3.2. Intrathecal administration and intrathecal microdialysis

The individual intrathecal concentration–time profiles of bupivacaine and lidocaine following intrathecal administration of an equimolar bupivacaine–lidocaine mixture are presented in Fig. 3. The values of the biopharmaceutic parameters are summarized in Table 2. The concentrations of bupivacaine and lidocaine in plasma were below the limits of detection during the intrathecal experiment. The C_{max}, AUC-csf-it and CL of lidocaine were very close to the corresponding

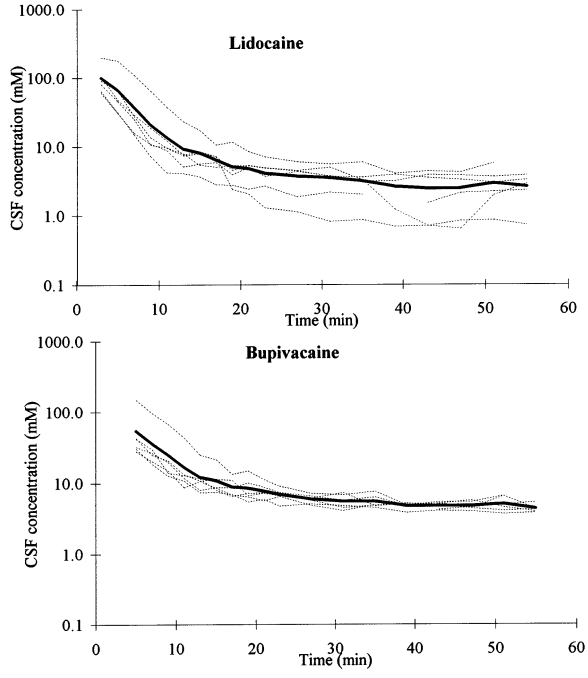

Fig. 3. Individual and mean CSF concentrations of bupivacaine (bottom) and lidocaine (top) after intrathecal administration of an equimolar bupivacaine–lidocaine mixture (0.2 μM) in rabbits.

Table 2
Intrathecal biopharmaceutic parameters (mean ± S.D.) of bupivacaine and lidocaine after intrathecal administration of an equimolar bupivacaine–lidocaine mixture (0.2 μM) in rabbits

		Bupivacaine	Lidocaine
C_{max}	(μg/ml)	61 ± 43	99 ± 52**
	(mM)	0.21 ± 0.15	0.42 ± 0.22
AUC-csf-it	(mM.min)	2.3 ± 1.2	2.8 ± 1.8
CL	(ml/min)	0.100 ± 0.031	0.087 ± 0.034*

* $P<0.05$.
** $P<0.01$.

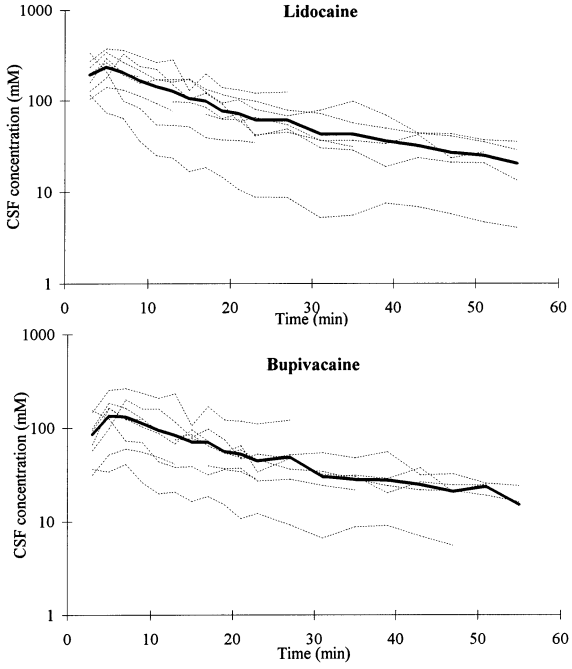

Fig. 4. Individual and mean CSF concentrations of bupivacaine (bottom) and lidocaine (top) after epidural administration of an equimolar bupivacaine–lidocaine mixture (6.9 μM) in rabbits.

Table 3
Intrathecal biopharmaceutic parameters (mean ± S.D.) of bupivacaine and lidocaine after epidural administration of an equimolar bupivacaine–lidocaine mixture (6.9 μM) in rabbits

		Bupivacaine	Lidocaine
C_{max}	(μg/ml)	154 ± 72	270 ± 94**
	(mM)	0.53 ± 0.25	1.14 ± 0.39
T_{max}	(min)	5.8 ± 1.5	4.8 ± 1.3
AUC-csf-epi	(mM min)	9.8 ± 5.5	17.3 ± 8.3**
K_a	(min^{-1})	0.762 ± 0.654	0.261 ± 0.092

** $P < 0.01$

values determined after separate administration of lidocaine. However, we found a dramatic decrease in C_{max} and AUC-csf-it values of bupivacaine compared with the separate intrathecal injection of bupivacaine ($C_{max} = 0.43 \pm 0.28$ mM, AUC-csf-it = 7.3 ± 3.6 mM min). Indeed, the intrathecal clearance of bupivacaine was higher than after separate administration of bupivacaine, the mechanism of this phenomenon remaining unexplained. It may be related to an increase of the diffusion from the intrathecal space to the epidural space and/or to a variation of the spinal nerves binding.

3.3. Intrathecal and plasma disposition after epidural administration

The individual intrathecal concentration–time profiles of bupivacaine and lidocaine after the epidural administration of an equimolar bupivacaine–lidocaine mixture are illustrated by the Fig. 4. The biopharmaceutic parameters are presented in Table 3. The mean total plasma C_{max} of bupicavaine and lidocaine were 0.71 ± 0.31 and 0.94 ± 0.40 μM, respectively. The corresponding T_{max} was 6.0 ± 2.6 min for both drugs. Compared with separate administration, this systemic resorption was slower and lower, especially for bupivacaine ($C_{max} = 2.23 \pm 1.25$ μM, $T_{max} = 1.6 \pm 1.5$ min). Thus, the mean CSF bioavailability of bupivacaine and lidocaine after simultaneous epidural administration was 12.3 and 17.9%, respectively. The mean CSF bioavailability of bupivacaine in the current study (12.3%) was higher than that of bupivacaine (5.5%) measured after its single administration while there was no change in the CSF bioavailability of lidocaine (17.9 vs. 17.7%) (Clément et al., 1999).

The fate of drug epidurally administered should result from different competitive processes summarized in Fig. 5: (1) transmeningeal transfert from the epidural to the intrathecal space; (2) distribution in the epidural fat; (3) systemic re-

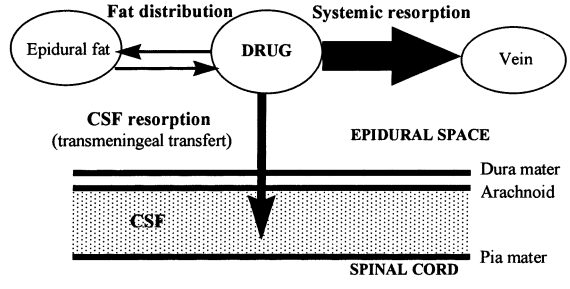

Fig. 5. Schematic representation of the competitive processes involved in the disposition of drugs administered in the epidural space.

sorption. The reduction in rate and extent in the systemic absorption might increase the intrathecal bioavailability of bupivacaine. Following epidural administration, the absorption rate constants (K_a) into the intrathecal space of bupivacaine and lidocaine were very close to the values obtained after the separate administration of each drug. Moreover, the K_a was around three times higher for bupivacaine compared with lidocaine, as found after separate administration (Clément et al., 1999). The lack of modification of K_a was not surprising since the transmeningeal transfert should result from a passive phenomenon. Because the K_a was not altered by the simultaneous administration of lidocaine, the difference in bupivacaine CSF bioavailability may result from a vascular effect of lidocaine reducing the systemic absorption of bupivacaine from the epidural space leading to an increase of its extent of absorption through meninges into CSF. Indeed, a large epidural dose of lidocaine induces a vasoconstriction of epidural veins (Myers and Heckman, 1989), that should lead to a reduction of the systemic resorption of bupivacaine which is the main process of elimination from the epidural space.

The increased CSF bioavailability of bupivacaine was unexpected since the administration of mixture containing bupivacaine and lidocaine has a shorter duration of effect than bupivacaine administered alone (Seow et al., 1982). However, the CSF disposition of local anesthetics results from different processes remaining yet unclear. Moreover, clinical studies measure pharmacodynamics effects such as duration of motor block, which may not be directly related to the intrathecal bioavailabilities.

This work has shown that the microdialysis technique should be a promising and useful tool to determine the local bioavailability and to study the disposition of drugs currently epidurally administered, such as opiates or such as drug delivery systems of local anesthetics (Le Corre et al., 1995; Dollo et al., 1998; Malinovsky et al., 1999), which are investigated to improve the clinical and toxicological features of these agents.

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