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## Effects of Fetal pH on Local Anesthetic Transfer across the Human Placenta

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**Background:** Fetal acidemia increases umbilical venous bupivacaine concentrations in the *in situ* rabbit model. The authors studied the effects of decreasing fetal pH on the rate of maternal to fetal (M→F) clearances of lidocaine, bupivacaine, 2-chloroprocaine, and antipyrine (a nonionic marker of placental transfer) across the isolated, dual perfused, human placental cotyledon.

**Methods:** Maternal to fetal clearances of bupivacaine, lidocaine, 2-chloroprocaine, and antipyrine were determined at fetal pH (7.4), during progressive fetal acidemia (pH 7.2→7.0→6.8), and after recovery to fetal pH 7.4 in experiments with both low protein state and in those with *in vivo* maternal and fetal protein-binding potentials.

**Results:** Placental transfer of all three agents increased linearly as the fetal pH decreased. Antipyrine transfer was unaffected. Clearance of lidocaine and bupivacaine, but not 2-chloroprocaine, returned to baseline when fetal pH was restored to 7.4. When maternal and fetal protein-binding potentials were increased, clearance at fetal pH 7.4 of bupivacaine, but not lidocaine, decreased significantly. During fetal acidemia, the transfer of both agents increased, but to a lesser extent than in the low protein concentration experiments.

**Conclusions:** Increasing the pH difference between maternal and fetal perfusates promotes M→F passage of unionized lido-

caine, bupivacaine, and 2-chloroprocaine. This likely results from an increased proportion of ionized local anesthetic in the acidic fetal perfusate and consequent widening of the M→F concentration gradient of the unionized form. Transfer of lidocaine and bupivacaine was limited by the maternal protein binding. (Key words: Anesthesia; obstetric. Anesthetics, local: bupivacaine; 2-chloroprocaine; lidocaine. Pharmacokinetics: ionic gradient; placental transfer.)

NUMEROUS investigators using different animal models have studied the effects of fetal acidemia on local anesthetic distribution across the placenta, demonstrating fetal accumulation as the fetal pH decreases.<sup>1-5</sup> All concluded that these fetal pH-induced changes in transplacental distribution are the result of the trapping of the ionic form of these weakly basic compounds within the fetus. This concept of "ion trapping" reflects the change in equilibrium between the unionized, highly diffusible parent compound and its poorly diffusible ionized salt, in accordance with the drug's pK<sub>a</sub> and the tissue pH. The greater the difference between the decreasing fetal pH and the agent's pK<sub>a</sub>, the greater the proportion of ionized to unionized compound (table 1) and therefore the greater the concentration of trapped ionic drug in the fetal circulation. "Ion trapping" is, therefore, the accepted explanation for fetal local anesthetic accumulation with fetal acidemia.

In a study using the *in vivo* perfused rabbit placenta, pH-dependent increases in maternal to fetal (M→F) transfer of bupivacaine and meperidine were identified.<sup>1</sup> The authors of this article suggested that fetal acidemia increased not only the maternal/fetal equilibrium ratio but also the rate of M→F transfer of bupivacaine and meperidine. Differences in placentation<sup>6</sup> and the arrangement of the maternal/fetal circulatory pattern<sup>7</sup> (countercurrent *vs* concurrent) between the rabbit and human placenta make direct extrapolation of these data to the human placenta difficult. To date, this question has not been studied using the human placenta.

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Table 1. Physicochemical Properties of Local Anesthetics

Local Anesthetic	pK <sub>a</sub>	Source
Lidocaine	7.8	Wood <sup>19</sup> (heptane/buffer)
Bupivacaine	8.1	Wood <sup>19</sup>
Chloroprocaine	8.2	Ralston and Sholder <sup>20</sup>

The isolated human placenta has been used for the study of local anesthetic transfer. The isolated human placenta has been used for the study of local anesthetic transfer. The isolated human placenta has been used for the study of local anesthetic transfer.

### Methods

**Placental Perfusion.** With Institutional Review Board approval, written informed consent was obtained from healthy pregnant women. Either vaginal or cesarean section was performed. Single cotyledon placenta were perfused with heparinized maternal and fetal arterial and venous blood.

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# Ala-Kokko T, Pienimäki M. Transfer of lidocaine and bupivacaine across the placenta. ANESTHESIOLOGY 1993; 79:1000-1002.

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**Table 1. Physicochemical Properties of Bupivacaine, Lidocaine, and 2-Chloroprocaine**

	Partition Coefficient*	Maternal Protein Binding†	Molecular Weight‡	pK <sub>a</sub> ‡	% Unionized at pH‡			
					7.4	7.2	7.0	6.8
Lidocaine	2.9	60-75	234	7.90	24	17	11	7
Bupivacaine	28.0	90-97	288	8.10	17	11	7	5
Chloroprocaine	0.14	—	307	8.70	4.8	3.1	2	1.2

\* Wood<sup>19</sup> (heptane/buffer, pH 7.4).† Wood.<sup>19</sup>‡ Ralston and Shnider.<sup>20</sup>

The isolated human placental perfusion model<sup>8</sup> has been used effectively to study the transfer of many substances, and recently was used to investigate anesthetic agents.<sup>||, #, \*\*9</sup> Steady state drug clearances can be measured using an open (nonrecirculating), dual perfusion arrangement of this model. By controlling local anesthetic concentrations on the maternal side of the circuit while altering fetal circuit pH, effects on steady state clearance can be assessed. Using the dual perfused human placental model, the effect of fetal perfusate pH on the steady state clearance rates of bupivacaine, lidocaine, and 2-chloroprocaine, three weakly basic local anesthetic agents commonly used in obstetrics were compared with the nonionic transfer marker antipyrine.

## Methods

### Placental Perfusions

With Institutional Review Board approval, and informed, written consent, term placentae were obtained from healthy parturients within 5-10 min after either vaginal or caesarean delivery. The dual perfused single cotyledon placental model was used as described previously.<sup>9</sup> The placenta was immediately perfused with heparinized Krebs-Ringer buffer (KRB). A fetal artery and vein supplying a single cotyledon

were cannulated with polyethylene infant feeding tubes (Sherwood Medical, St Louis, MO) and perfused with KRB. After ensuring the integrity of the circulation, the placental cotyledon was placed in a plexi-glass chamber, maternal side up. The intervillous space was perfused using three blunt-tipped, 19-gauge needles inserted through the decidual plate in the region of the maternal surface blanched by the blood-free media already perfusing the fetal circuit. All experiments were performed using the open (single pass or nonrecirculating) method and tested the transfer of lidocaine, bupivacaine, 2-chloroprocaine, and antipyrine in the M→F direction. Maternal and fetal circulatory temperature were maintained at 37°C. The maternal flow rate was maintained at 12-15 ml/min, which produced perfusion pressures of 25-35 mmHg. The fetal flow rate was adjusted to provide fetal perfusion pressures of 65-75 mmHg, which resulted in flow rates of 1.5-3.0 ml/min. Pressure, pH, and temperature were electronically recorded using a specific software package (Anesthesiology Research Department, Vanderbilt University) for the duration of the experiment. Throughout the experiment, 2-ml samples were concurrently obtained from the maternal reservoir and fetal venous outflow at 15-min intervals. Maternal and fetal samples were immediately placed in tubes that contained 0.2 mg sodium fluoride.

Antipyrine (10 µg/ml) was added to the maternal circuit in each experiment to act as a nonionic marker of transfer. Local anesthetic clearances were expressed as a ratio of the corresponding antipyrine clearance to allow for individual placental preparation variations.

### Specific Experiments

Three groups of experiments were designed to elucidate the role of three possible factors in the phenomenon of increased local anesthetic transfer during acid-

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emia, namely, enhanced gradient of unionized drug, maternal and fetal protein binding, and tissue permeability changes.

The effect of differential gradient of the unionized forms of the test agents were studied in 14 experiments. To minimize protein binding, the maternal and fetal perfusate consisted of KRB supplemented with 0.2 g/100 ml human albumin and 150 mg/100 ml glucose. In these experiments, 1  $\mu$ g/ml bupivacaine and 5  $\mu$ g/ml lidocaine were administered concurrently into the maternal circuit. In eight of these experiments, 5  $\mu$ g/ml 2-chloroprocaine was studied along with lidocaine and bupivacaine. Clearance for each agent was initially calculated at a pH of 7.4 in both circuits and again while fetal perfusate pH was changed every 30 min to 7.2, 7.0, 6.8, and back to 7.4.

In another three experiments, the effect of maternal protein binding and pH on lidocaine and bupivacaine transfer were studied using fresh frozen plasma (FFP) for the maternal perfusate and KRB with 4.0 g/100 ml human albumin as the fetal perfusate. In these experiments, the original solution of 5  $\mu$ g/ml lidocaine and 1  $\mu$ g/ml bupivacaine was used. 2-chloroprocaine was not used because of possible metabolism by the FFP. Maternal to fetal clearances of the test agents were studied at fetal pH 7.4 (45 min), 6.8 (60 min), and 7.4 (60 min).

In three additional KRB experiments, any possible pH-related change in placental permeability was addressed. The gradient of unionized drug was equalized between the fetal and maternal circuits by monitoring the test agent's clearance for 1 h with the pH of both the maternal and fetal circuits at 7.4 and afterwards for another hour with both at pH 6.8.

In all experiments, the pH perturbations were achieved by the addition of dilute hydrochloric acid or sodium bicarbonate, and the adjusted pH was maintained within a narrow range, using carbon dioxide flow and continued equilibration with 21% O<sub>2</sub>.

#### High Performance Liquid Chromatographic Analysis

Bupivacaine, lidocaine, 2-chloroprocaine, and antipyrine concentrations were concurrently measured in perfusate samples using a modified high performance liquid chromatographic method, as described by Ha *et al.*<sup>10</sup> Briefly, the internal standard (25  $\mu$ l butorphanol of 25  $\mu$ g/ml aqueous solution) was added to standard and 0.5-ml experimental samples made basic by the addition of 0.2 ml 2N NaOH and extracted with ethyl

ether. The ether phases were vaporized to dryness under N<sub>2</sub>, and the residue dissolved in 150  $\mu$ l mobile phase. These extracted samples were then analyzed by high performance liquid chromatography. The chromatographic system (Waters Associates, Milford, MA) consisted of a 501 pump, WISP 712, and a 486 UV detector (210 nm). The high performance liquid chromatographic column,  $\mu$ Bondapak C18 (Waters Associates, Milford, MA), was equilibrated at 1 ml/min with acetonitrile and 0.05 M Na<sub>2</sub>HPO<sub>4</sub>, pH 5.8 (30:70 v/v). Local anesthetic and antipyrine concentrations in perfusate samples were ascertained from peak-height ratios with known concentrations of each agent. Concentration curves for the four test agents were linear from 0.1  $\mu$ g to 10.0  $\mu$ g/ml. The determination of bupivacaine, lidocaine, 2-chloroprocaine, and antipyrine exhibited an interday coefficient of variation of 4.1, 4.4, 4.0, and 4.4, respectively.

#### Calculations

Because the purpose of this investigation was to elucidate the movement of drug from the maternal circulation to the fetal circulation during the period of acidemia, clearance was used as the means of measurement of drug transfer. In an open perfusion, a steady state M→F clearance is attained rapidly, and can be calculated using the formula:

$$Cl_{m \rightarrow f} = (C_{FV} \times Q_F) / C_{MA}$$

where C<sub>FV</sub> = drug concentration in fetal venous effluent, C<sub>MA</sub> = drug concentration in the "maternal artery" (maternal reservoir), and Q<sub>F</sub> = fetal perfusate flow rate.<sup>8</sup> Transfer ratio was calculated by dividing the local anesthetic clearance by the corresponding antipyrine clearance. Transfer ratio was used to compare clearance rates between the KRB and FFP experiments due to changes in baseline clearance values as a result of the increased maternal protein binding.

#### Statistics

Mean clearances observed for each agent at varying fetal pH were examined using one-way repeated measures analysis of variance. Pairs-wise comparisons of clearance values for pH 7.2, 7.0, and 6.8 with control (pH 7.4) on any agent in which a significance was detected by repeated measures analysis of variance using the Bonferroni corrected *t* test. Lidocaine and bupivacaine clearance, KRB versus FFP, was compared using unpaired *t* test. Lidocaine and bupivacaine clear-

Table 2. Clearance

Lidocaine (n = 14)
Bupivacaine (n = 14)
Chloroprocaine (n = 14)
Antipyrine (n = 14)

\* Return of pH to baseline  
 † P ≤ 0.05 versus control  
 ‡ P ≤ 0.05 by t ratio of

ance, KRB versus FFP  
 † test. Linear regression  
 correlation between fetal  
 cent unionized drug  
 Data are expressed as mean  
 was considered significant

#### Results

##### Gradient Experiments

With equal, low fetal circuit bupivacaine and antipyrine concentrations introduced into the fetal circuit, observed at the test pH. All four agents had clearances that were pH dependent. The local anesthetic agents had clearances that increased to 2, 7, and 7.4. No such alteration was observed. A statistically significant difference was shown between the agent and the pH. In addition, a close relationship was calculated between the clearances compared with the control. The clearance for each agent returned to near baseline values at pH 7.4. In contrast, the clearances returned to baseline

##### Protein Binding

When the protein binding of the fetal perfusate was enhanced, the clearance of bupivacaine

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**Table 2. Clearance Data at Progressively Lower Fetal Perfusate pH**

	Clearance (ml · h <sup>-1</sup> · g <sup>-2</sup> ) at pH					Correlation Coefficient
	7.4	7.2	7.0	6.8	7.4*	
Lidocaine (n = 14)	4.86 ± 0.62	5.72 ± 0.66†	6.68 ± 0.73	7.40 ± 0.88†	5.12 ± 1.12	0.992‡
Bupivacaine (n = 14)	3.89 ± 0.65	4.90 ± 0.68†	5.76 ± 0.72†	6.32 ± 0.85†	4.21 ± 0.93	0.999‡
Chloroprocaine (n = 8)	4.44 ± 0.79	5.30 ± 0.83†	6.55 ± 0.94†	7.75 ± 1.03†	6.77 ± 1.82†	0.994‡
Antipyrine (n = 14)	5.33 ± 0.67	5.43 ± 0.60	5.93 ± 0.64	5.83 ± 0.80	5.13 ± 0.69	0.771

\* Return of pH to baseline following completion of protocol.

†  $P \leq 0.05$  versus control (7.4) by Bonferroni corrected Student's paired *t* test.

‡  $P \leq 0.05$  by *t* ratio of the regression coefficient to its standard error.

ance, KRB versus FFP, was compared using unpaired *t* test. Linear regression analysis was used to assess correlation between the pH of the fetal circuit or percent unionized fraction and the clearances observed. Data are expressed as mean ± SEM. *P* value ≤ 0.05 was considered significant.

## Results

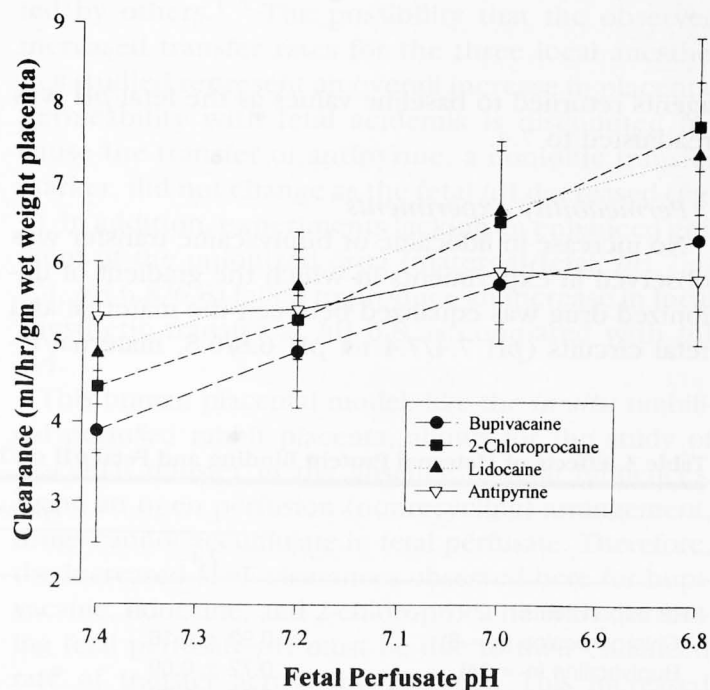
### Gradient Experiments

With equal, low protein binding in both maternal and fetal circuits, bupivacaine, lidocaine, 2-chloroprocaine, and antipyrine crossed the placenta rapidly after their introduction into the maternal circuit. Clearance values observed at the tested pH stages are presented in table 2. All four agents studied exhibited similar baseline clearances at pH 7.4. A linear increase in M→F clearance (transfer) was observed for each of the three local anesthetic agents as the fetal perfusate pH was decreased to 7.2, 7.0, and 6.8 (fig. 1). Antipyrine exhibited no such alteration in M→F clearance when pH was decreased. A statistically significant correlation was shown between the clearance of each local anesthetic agent and the pH of the fetal perfusate (table 2). In addition, a close relation was demonstrated when the calculated percent of unionized drug (table 1) was compared with the clearance of each test drug (fig. 2). The clearance for lidocaine and bupivacaine returned to near baseline when fetal pH was readjusted to pH 7.4. In contrast, 2-chloroprocaine clearance failed to return to baseline.

### Protein Binding Experiments

When the protein binding potential of the maternal perfusate was enhanced, baseline (fetal pH 7.4) M→F clearance of bupivacaine was significantly reduced, as

indicated by the associated transfer ratios (KRB, 0.72 ± 0.09 vs. FFP, 0.29 ± 0.06,  $P \leq 0.05$ ). Conversely, lidocaine clearance was unaffected by increasing the maternal protein binding (KRB, 0.89 ± 0.09 vs. FFP, 0.80 ± 0.07, not significant). The transfer ratios of both agents increased significantly during the periods of acidemia, albeit to a lesser degree than that seen in the KRB experiments (table 3). The transfer ratio of both



**Fig. 1.** Plot illustrating the effect of fetal perfusate pH on the maternal-to-fetal clearance of bupivacaine, lidocaine, 2-chloroprocaine, and antipyrine. Steady state drug transfer was established at a pH of 7.4 in both the maternal and fetal circuits, after which the fetal perfusate pH was changed to 7.2, 7.0, 6.8, and back to 7.4 in 30-min intervals. A linear increase in clearance was observed for each of the local anesthetic agents throughout the range of pHs studied, but not for antipyrine. Data expressed as mean ± SEM.

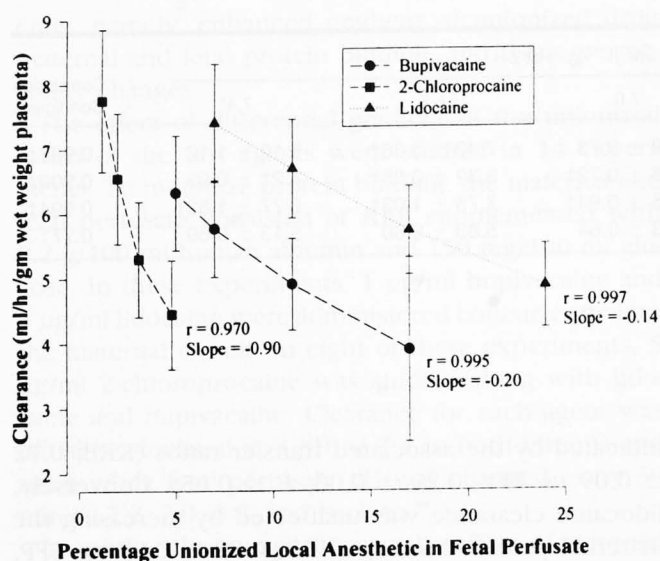


Fig. 2. Plot illustrating the relation of the calculated fraction of the unionized bupivacaine, lidocaine, and 2-chloroprocaine in the fetal perfusate on maternal-to-fetal clearance. Clearance was shown to be inversely and linearly related to the unionized fraction of drug in the fetal circuit. The correlation coefficient,  $r$ , and slope for a regression line for each agent is noted below each curve. Data expressed as mean  $\pm$  SEM.

agents returned to baseline values as the fetal  $pH$  was readjusted to 7.4.

#### Permeability Experiments

No increase in lidocaine or bupivacaine transfer was observed in experiments in which the gradient of unionized drug was equalized between the maternal and fetal circuits ( $pH$  7.4/7.4 vs.  $pH$  6.8/6.8, maternal/fetal), demonstrated by the unchanged clearance ratios for bupivacaine ( $0.72 \pm 0.03$  vs.  $0.68 \pm 0.04$ ), lidocaine ( $0.93 \pm 0.02$  vs.  $0.93 \pm 0.03$ ), and 2-chloroprocaine ( $1.03 \pm 0.09$  vs.  $0.99 \pm 0.06$ ) at  $pH$  7.4 and  $pH$  6.8. Maternal to fetal clearance of antipyrine ( $5.6 \pm 0.6$ ,  $5.8 \pm 0.7$ ,  $pH$  7.4/6.8, respectively) was similar at both  $pH$  intervals.

**Viability Parameters**  
Lactate production was  $0.165 \pm 0.06$   $\mu\text{mol}/\text{min}/\text{g}$ , and glucose consumption measured  $0.11 \pm 0.05$   $\mu\text{mol}/\text{min}/\text{g}$ , which were similar to values associated with oxygenated systems as observed by others.<sup>11-13</sup> No significant fluid shift was observed from the maternal circulation to the fetal circuit, reflecting system integrity without leakage of perfusate throughout the perfusion period.

#### Discussion

In 1976, Brown *et al.*<sup>14</sup> reported four cases in which acidemic newborns ( $pH$  7.0-7.02) were shown to have high umbilical vein/maternal vein concentration ratios of mepivacaine and lidocaine. Since then, others investigated the influence of fetal  $pH$  on the distribution of local anesthetics across the placenta. Using animal models,<sup>1-5</sup> researchers related these increased umbilical vein concentrations of weakly basic drugs to the  $pH$  of the blood perfusing the umbilical vessels. These investigations clearly demonstrate that widening the  $pH$  gradient between the maternal and fetal circulations influences the M $\rightarrow$ F distribution of local anesthetics across the placenta. Some possible explanations for this distribution have been suggested.<sup>15,16</sup> A possible mechanism is the ionized form of the drug, which is more readily distributed to fetal tissue, labeled fetal tissue, and fetal tissue.<sup>15,16</sup> Recent studies have investigated the distribution of mepivacaine and lidocaine across the placenta. Only the maternal side was affected by fetal acidemia. Ethical concerns prevented the use of direct fetal anesthesia to detect directly fetal anesthesia. Contractions reported by humans.<sup>14</sup> However, human placental studies showed that  $pH$  differences appear to play a role in the weakly basic local anesthetic distribution. The concept that the M $\rightarrow$ F transfer is affected by the work of Biehl and coworkers. With both maternal and fetal  $pH$  constant, these studies showed that the  $pH$  gradient was the cause of the increased bupivacaine transfer. The work of Biehl and coworkers showed that the  $pH$  gradient was the cause of the increased bupivacaine transfer. The work of Biehl and coworkers showed that the  $pH$  gradient was the cause of the increased bupivacaine transfer.

Table 3. Effects of Maternal Protein Binding and Fetal  $pH$  on Local Anesthetic/Antipyrine Clearance Ratios

	Antipyrine Clearance Ratios at $pH$			% Change $\ddagger$
	7.4	6.8	7.4	
KRB experiments				
Chloroprocaine (n = 8)	$0.92 \pm 0.16$	$1.67 \pm 0.31^*$	$1.32 \pm 0.35^*$	82
Bupivacaine (n = 14)	$0.72 \pm 0.09$	$1.26 \pm 0.11^*$	$0.91 \pm 0.18$	75
Lidocaine (n = 14)	$0.89 \pm 0.09$	$1.25 \pm 0.10^*$	$0.94 \pm 0.18$	40
FFP experiments				
Bupivacaine (n = 3)	$0.29 \pm 0.06^\dagger$	$0.35 \pm 0.06^*$	$0.28 \pm 0.05$	21
Lidocaine (n = 3)	$0.80 \pm 0.07$	$1.01 \pm 0.09^*$	$0.81 \pm 0.05$	26

\*  $P \leq 0.05$  versus control ( $pH$  7.4) clearances by Bonferroni corrected Student's paired  $t$  test.

$\dagger P \leq 0.05$  versus baseline bupivacaine clearances in KRB experiments by paired  $t$  test.

$\ddagger$  As compared with initial baseline clearances at  $pH$  7.4.

KRB = Krebs-Ringer bicarbonate; FFP = fresh frozen plasma.

tics across the placenta. Some possible explanations for this distribution have been suggested.<sup>15,16</sup> A possible mechanism is the ionized form of the drug, which is more readily distributed to fetal tissue, labeled fetal tissue, and fetal tissue.<sup>15,16</sup> Recent studies have investigated the distribution of mepivacaine and lidocaine across the placenta. Only the maternal side was affected by fetal acidemia. Ethical concerns prevented the use of direct fetal anesthesia to detect directly fetal anesthesia. Contractions reported by humans.<sup>14</sup> However, human placental studies showed that  $pH$  differences appear to play a role in the weakly basic local anesthetic distribution. The concept that the M $\rightarrow$ F transfer is affected by the work of Biehl and coworkers. With both maternal and fetal  $pH$  constant, these studies showed that the  $pH$  gradient was the cause of the increased bupivacaine transfer. The work of Biehl and coworkers showed that the  $pH$  gradient was the cause of the increased bupivacaine transfer.

Using a perfused placental transfer model (also a salt of a weakly basic drug), the  $pH$  was reduced and the effect of bupivacaine and mepivacaine was demonstrated.

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tics across the placenta, as reflected in an increased maternal/fetal concentration ratio. Several mechanisms have been suggested to explain this phenomenon. Some possible explanations include: Decreased local anesthetic clearance by the fetus; changes in tissue distribution<sup>15,16</sup>; and the trapping of the poorly diffusible ionized form of the local anesthetic in acidemic fetal tissue, labeled "ion trapping."

Recent studies of perfused rabbit placentae *in situ* investigated the fetal pH effects on bupivacaine and meperidine transfer. The resultant data suggest that not only is the maternal/fetal equilibrium ratio increased by fetal acidemia, but so is the rate of drug transfer.<sup>1</sup> Ethical concerns and inaccessibility make it impossible to detect directly whether increased transfer of local anesthetics contribute to their increased concentrations reported clinically with fetal acidemia in humans.<sup>14</sup> However, by using the *in vitro* dual perfused human placental model in the current study, we showed that pH-dependent influences on M→F transfer appear to play a role in altering the distribution of weakly basic local anesthetics across the human placental cotyledon.

The concept that fetal acidemia induces an increase in the M→F transfer of local anesthetics was first suggested by the work of Biehl *et al.*<sup>2</sup> using pregnant ewes. With both maternal and fetal circulations instrumented, Biehl and coworkers found that fetal arterial concentrations of lidocaine increased with decreasing fetal pH when maternal lidocaine concentrations were held constant. These workers concluded that "ion trapping" was the causal factor, but that increased lidocaine transfer was also a possible factor. Because fetal blood samples were obtained from the femoral artery rather than the umbilical vein, their results probably reflect the function of the materno-placento-fetal unit as a whole. Therefore, fetal mechanisms that affect plasma local anesthetic concentrations cannot be excluded. In a later publication, these investigators reported similar pH-related changes in the placental transfer of bupivacaine. They concluded that bupivacaine's increased maternal protein binding potential did not abolish the effects of fetal acidemia as might be expected.<sup>3</sup>

Using a perfused *in situ* rabbit placenta, Gaylard and coworkers<sup>1</sup> demonstrated a pH-dependent increase in placental transfer of both bupivacaine and meperidine (also a salt of a weak base) when the umbilical perfusate pH was reduced to 7.0. Placental clearance of bupivacaine and meperidine increased 160% and 80%, re-

spectively, when the fetal perfusate was acidified, but returned to baseline after the pH was normalized back to 7.4. The authors observed no such alteration in the rate of transfer of antipyrine in their study. In addition, a decrease in maternal uterine perfusion produced by a withdrawal of blood from the rabbit doe to effect a 65% reduction in mean maternal arterial pressure induced an increase in bupivacaine clearance. In contrast, both meperidine and antipyrine clearances decreased with hypovolemic hypotension. Maternal/fetal equilibrium ratio, however, was unaffected by maternal placental hypoperfusion in the absence of fetal acidemia. The authors suggested that bupivacaine's high lipid solubility and protein binding might be the major factors involved in enhancing transfer during maternal hypotension. However, no explanation for the increased clearance of bupivacaine with fetal acidemia was offered.

In the current investigation, increases in M→F clearances of bupivacaine, lidocaine, and 2-chloroprocaine were demonstrated in isolated, dual-perfused human placental cotyledons when fetal perfusate pH values were decreased to levels similar to those studied by others.<sup>1-5</sup> The possibility that the observed increased transfer rates for the three local anesthetics studied represent an overall increase in placental permeability with fetal acidemia is discounted because the transfer of antipyrine, a nonionic transfer marker, did not change as the fetal pH decreased (fig. 1). In addition, experiments lacking an enhanced gradient of the unionized drug (maternal/fetal, pH 7.4/7.4, pH 6.8/6.8) failed to produce an increase in local anesthetic transfer at pH 6.8 as compared with pH 7.4.

This human placental model, like the *in situ* umbilical perfused rabbit placenta, allows for the study of placental transfer in the absence of fetal influences. Using an open perfusion (nonrecycling) arrangement, drugs cannot accumulate in fetal perfusate. Therefore, the increased M→F clearances observed here for bupivacaine, lidocaine, and 2-chloroprocaine with decreasing fetal perfusate pH must be due to their enhanced rate of transfer across the placenta. This increased transfer rate is most likely the result of pH-induced changes in the maternal intervillous:fetal sinusoid gradient for the unionized drug. It can be assumed that, as the local anesthetics cross from maternal to a more acidic fetal blood, a greater proportion of each agent will be protonated to the ionized form in proportion to their pK<sub>a</sub> values. If less unionized drug is present in

the fetal circulation because of an acidemic environment, a greater maternal:fetal gradient of undissociated local anesthetic will be maintained throughout the time the fetal blood is in apposition with the maternal intervillous blood, thereby increasing transfer. This hypothesis is supported by the highly significant correlation observed in the current study between local anesthetic drug clearance and calculated percentage of unionized drug in the fetal circulation (fig. 2).

Because the test agents are believed to cross the human placenta by passive diffusion,<sup>1</sup> their individual M→F clearance rates should not be affected by the presence of multiple test agents. Maternal to fetal placental clearance of various combinations of agents were studied previously in animal and human placental models, with no competitive inhibition observed in their transfer process.<sup>9,17</sup> Bupivacaine placental transfer was shown to be unaffected by the addition of increasing concentrations (as much as 20 times higher) of mepivacaine, an analog of bupivacaine.<sup>9</sup> The effect of protein binding and fetal flow on the placental transfer of concurrently administered bupivacaine, lidocaine, and meperidine were determined in perfused rabbit placenta with success.<sup>17</sup> In addition, these local anesthetic agents are used clinically, often in various combinations, to provide better analgesia. Therefore, combinations of test agents were used in the perfused placenta experiments because this procedure provided the best opportunity to study the transfer of the test agents under the identical acidemic conditions.

The protein bindings potentials of these agents vary widely (table 1). Therefore, this investigation was performed both in a minimal protein perfusate (0.2 mg/100 ml) and in a perfusate (FFP) that allows "physiologic" protein binding similar to those observed *in vivo*. In the KRB perfused placental experiments where maternal protein binding was not a factor, the M→F clearances for bupivacaine, lidocaine, and 2-chloroprocaine at the baseline fetal pH of 7.4 were all similar. However, in agreement with a prior publication,<sup>9</sup> baseline (fetal pH 7.4) bupivacaine clearance was markedly reduced by substitution of FFP for KRB as the maternal perfusate. No significant reduction was observed with lidocaine. This decrease in baseline clearance for bupivacaine as opposed to that of lidocaine is consistent with their relative degrees of protein binding (table 1). As with the studies of Pickering *et al.*,<sup>3</sup> a significant increase in bupivacaine transfer was observed with fetal acidemia, even with the greatly increased maternal protein binding of the FFP. Because

the increase in local anesthetic clearance during the acidemic period was less in the FFP experiments compared with the KRB experiments, it is demonstrated that maternal protein binding local anesthetic transplacental movement during fetal acidemia. These pH-associated changes in the FFP experiments may more accurately represent those anticipated to occur *in vivo*, where the natural physiologic forces of maternal protein binding are at work.

Because these agents were studied simultaneously in the same placentae, distribution of maternal and fetal blood flow was similar for each of the local anesthetics investigated. Therefore, a factor or factors other than protein binding and maternal/fetal blood flow distribution must have played the major role in influencing the rate of drug transfer under these study conditions.

One possible factor could be the damage to the fetal intravillous vasculature induced by fetal acidemia (pH 6.8). Lack of such trauma is evidenced by the return to baseline clearance values observed for both lidocaine and bupivacaine after the acidemic portion of the investigation. Chloroprocaine's failure to return to baseline clearance values is unexplained.

Information gained from this investigation strongly supports the existence of a pH-dependent alteration in M→F clearance rates of weakly basic agents within the human placenta, much like that found previously within the rabbit placenta.<sup>1</sup> This increase in the transfer of local anesthetics in conjunction with "ion trapping" probably contributes to the elevated fetal levels of these agents when the fetus is compromised by acidemia. Based on the results of this investigation, the increase in drug transfer *in vivo* induced by fetal acidemia also may be limited by the agent's maternal protein binding. Despite this protective influence, local anesthetic concentrations in the fetal unit could still be elevated to a critical degree. This observation could be of importance to the safety of the preterm infant because investigators demonstrated increased sensitivity of the asphyxiated fetus to the potentially deleterious effects of local anesthetics on the fetus.<sup>4,5,18</sup> Based on these assumptions, in situations where the preterm fetus is likely to be acidemic, all efforts must be taken to minimize the maternal concentrations of local anesthetics, because fetal concentrations mirror those of the mother. The use of bupivacaine might be preferred to that of lidocaine, because of that agent's high degree of maternal protein binding.

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