where γ is the ratio of the concentration of albumin-bound spirolactone to that of albumin, D_f is the concentration of unbound spirolactone at dialysis equilibrium, K is the binding constant at equilibrium, and n is the number of a single type of binding sites. The previous investigators (7) apparently obtained linear plots of γ/D_f *versus* γ for several spirolactones. The concentrations of spirolactone used (7) appear to have been ~30–800 times higher than the peak serum concentrations of I and its metabolites detected in human males given a 200-mg oral dose (10).

The observed absence of linearity, together with the complexity of the Scatchard plots for I and II, strongly suggests diverse protein–ligand binding characteristics. Such deviations were reported to be due to cooperative ligand interaction, multiple-contact binding sites, or nonequivalent binding sites (20). Inspection of the Scatchard plots reveals several points of inflection, as well as both concave and convex curvatures at varying concentrations of bound I and II. Such patterns generally are the results of different types of binding sites, each exhibiting cooperative character. Furthermore, individual binding sites for I and II may overlap and also may contribute to the diverse binding pattern observed. Although no absolute conclusion can be drawn regarding which phenomenon dominates the binding pattern of I and II for human serum albumin, a single model of independent, equivalent binding is not applicable for the spirolactones studied.

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Lidocaine Pharmacokinetics in Pregnant and Nonpregnant Sheep

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Abstract □ Lidocaine disposition kinetics were studied in the pregnant ewe following 0.5-, 1.0-, and 2.0-mg/kg iv bolus doses and in the nonpregnant ewe following a 1.0-mg/kg iv bolus dose. Arterial blood was assayed for lidocaine by GLC. The blood lidocaine concentration–time curves were computer fitted to a two-compartment open model. In the pregnant ewe, the total body clearance of lidocaine (38 ml/min/kg) remained constant with increasing dose and was correlated linearly with preinjection cardiac output. The apparent volume of distribution of the central compartment apparently increased with increasing dose. The half-life of the postdistributive phase and the volumes of distribution at steady state and during the postdistributive phase increased as the dose was increased from 0.5 to 1.0 mg/kg. These observations suggest dose-related distribution of lidocaine in the pregnant ewe. The total body clearance of lidocaine in the pregnant ewe was not different from that in the nonpregnant ewe after 1.0-mg/kg doses; however, the volumes of distribution of the central compartment at steady state and during the postdistributive phase and the half-life of the postdistributive phase were greater in the pregnant ewe. The greater total body clearance for lidocaine in sheep as compared to humans is consistent with the greater hepatic blood flow in sheep; calculated hepatic extraction ratios for sheep are similar to hepatic extraction ratios for humans.

Keyphrases □ Lidocaine—pharmacokinetics, pregnant and nonpregnant sheep □ Pharmacokinetics—lidocaine, pregnant and nonpregnant sheep □ Anesthetics—lidocaine, pharmacokinetics, pregnant and nonpregnant sheep

Few literature reports describe drug disposition kinetics in the pregnant individual, but drug pharmacokinetics during pregnant and nonpregnant states may differ markedly. Alterations in drug disposition kinetics could be related to maternal changes, including blood and/or tissue binding of the drug, changes in the rates or distribution of blood flow, and drug metabolism changes. More importantly, drug kinetics during pregnancy may be altered by the addition of the fetal–placental unit with its inherent abilities to distribute, bind, metabolize, and clear drugs.

Lidocaine disposition kinetics at three doses in the pregnant ewe are presented in this report, and a comparison is made with lidocaine disposition kinetics in the nonpregnant ewe.

EXPERIMENTAL

Pregnant and nonpregnant pure or crossbred Suffolk ewes, 64.6 ± 11.3 (mean ± *SD* on the day of surgery) and 66.7 ± 11.9 kg, respectively, were obtained locally. Pregnant ewes were studied from Day 137 to Day 143 of gestation (full term 147–150 days). The ewes were catheterized during sterile surgery under general endotracheal anesthesia, using halothane and oxygen with controlled mechanical ventilation.

Polyethylene catheters were placed into the femoral artery and femoral vein and advanced to the abdominal aorta and inferior vena cava, respectively. The femoral artery catheter was used for continuous blood pressure and heart rate monitoring, and the femoral vein catheter was used for lidocaine administration. A central venous catheter was inserted *via* percutaneous puncture into the external jugular vein and positioned in the right atrium. This catheter was used for the injection of indocya-

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Table I—Estimates of Pharmacokinetic Constants for Individual Pregnant Ewes

Dose, mg/kg	Subject	Estimated Constants and Coefficients of Variation[a] A, μg/ml	α, min^{-1}	B, μg/ml	β, min^{-1}	Measures of Fit r^2	Correlation Coefficient
0.5	1	1.21 (0.286)	0.553 (0.174)	0.190 (0.192)	0.0169 (0.154)	0.997	0.989
	2	1.71 (0.195)	0.549 (0.125)	0.130 (0.150)	0.0174 (0.109)	0.999	0.997
	3	0.993 (0.165)	0.445 (0.112)	0.161 (0.111)	0.0187 (0.118)	0.999	0.971
	4	1.42 (0.114)	0.331 (0.078)	0.166 (0.081)	0.0130 (0.092)	1.000	0.993
	5	1.24 (0.162)	0.385 (0.102)	0.111 (0.123)	0.0176 (0.108)	1.000	0.985
	Average	1.31 (0.183)	0.453 (0.124)	0.152 (0.133)	0.0167 (0.120)		
1.0	1	3.02 (0.225)	0.415 (0.174)	0.181 (0.166)	0.0128 (0.203)	0.999	0.994
	2	1.07 (0.197)	0.282 (0.140)	0.159 (0.127)	0.0125 (0.168)	0.999	0.987
	3	1.69 (0.113)	0.531 (0.072)	0.317 (0.061)	0.00954 (0.084)	0.998	0.993
	4	1.73 (0.294)	0.282 (0.221)	0.144 (0.208)	0.0107 (0.411)	0.998	0.993
	Average	1.88 (0.212)	0.378 (0.141)	0.201 (0.124)	0.0114 (0.217)		
2.0	1	4.31 (0.150)	0.368 (0.097)	0.616 (0.114)	0.0161 (0.093)	1.000	0.965
	2	1.80 (0.171)	0.131 (0.121)	0.329 (0.111)	0.00797 (0.163)	0.998	0.936
	3	2.44 (0.121)	0.199 (0.078)	0.566 (0.081)	0.0100 (0.095)	0.999	0.967
	4	4.49 (0.157)	0.337 (0.097)	0.435 (0.124)	0.0170 (0.106)	1.000	0.957
	5	2.45 (0.089)	0.205 (0.058)	0.443 (0.063)	0.0160 (0.049)	1.000	0.990
	Average	3.10 (0.140)	0.248 (0.090)	0.478 (0.098)	0.0134 (0.094)		

[a] Numbers in parentheses are coefficients of variation.

Table II—Estimates of Pharmacokinetic Constants for Individual Nonpregnant Ewes

Subject	Estimated Constants and Coefficients of Variation[a] A, μg/ml	α, min^{-1}	B, μg/ml	β, min^{-1}	Measures of Fit r^2	Correlation Coefficient
1	3.34 (0.208)	0.473 (0.146)	0.136 (0.174)	0.0200 (0.110)	1.000	0.907
2	7.20 (0.187)	0.625 (0.099)	0.330 (0.123)	0.0126 (0.143)	1.000	0.985
3	2.16 (0.100)	0.216 (0.058)	0.395 (0.076)	0.0199 (0.050)	1.000	0.994
Average	4.23 (0.177)	0.438 (0.109)	0.287 (0.109)	0.0175[b] (0.095)		

[a] Numbers in parentheses are coefficients of variation. [b] Statistically significant ($p < 0.05$) difference between nonpregnant and pregnant ewes (Table I) using the Student t test.

nine green[1] in the cardiac output determination by a dye dilution technique.

An arterial catheter was placed through a branch of the carotid artery and advanced centrally to obtain arterial blood for blood gas, cardiac output, and lidocaine determinations. Arterial rather than venous blood was chosen for lidocaine determination because of expected arterial–venous lidocaine concentration differences resulting from tissue drug uptake (1, 2). The arterial lidocaine concentration better reflects presentation of the drug to well-perfused vital organs, especially during drug distribution. A laparotomy was performed on the pregnant ewe for placement of a fetal arterial catheter for obtaining fetal blood samples for blood gas and lidocaine determinations and for monitoring fetal heart rate and blood pressure.

All catheterizers were filled with heparinized saline and either tunneled subcutaneously to the flank or wrapped securely around the neck or hindlimb. The animal was allowed a minimum of 24 hr to recover from anesthesia and surgery before studies were initiated. For all animals, the heart rate, blood pressure, and blood gases were used as measures of well-being during surgery and subsequent studies. The results of these physiological determinations and the fetal blood lidocaine levels will not be presented in this report.

On experimental days, the animal was placed in a sheep-restraining cage and measurements were taken during a 30-min control period to produce baseline blood gas[2], blood pressure, and heart rate[3] data. Cardiac output was measured three times during the control period. Following the control period, lidocaine hydrochloride[4] was injected intravenously over 10 sec at one of three doses (0.5, 1.0, or 2.0 mg/kg) to the pregnant ewe or at one dose (1.0 mg/kg) to the nonpregnant ewe. All lidocaine doses are expressed as amounts of lidocaine hydrochloride administered. In the pregnant animal, the lidocaine doses were administered randomly on consecutive days following surgery to quantitate possible lidocaine pharmacokinetic variations as a function of postsurgical time.

No systematic variations were apparent in lidocaine pharmacokinetics or in the heart rate, blood pressure, or blood gases measured on consecutive days following surgery. Arterial blood samples were collected at 1, 2, 5, 10, 15, 20, 30, 45, 60, 90, 120, and 150 min following lidocaine injection and were stored frozen in heparinized tubes until subsequent determination of whole blood lidocaine by a GLC method having a coefficient of variation of <5% over the concentration range studied (3). All lidocaine concentrations and pharmacokinetic data were expressed in terms of lidocaine base. Four of the 14 pregnant ewe studies were performed on ewes with twin fetuses. No significant pharmacokinetic differences were found between the pregnant ewes carrying one fetus and those carrying two fetuses.

Blood lidocaine concentration–time curves for each ewe were described adequately by the biexponential equation:

$$C = Ae^{-\alpha t} + Be^{-\beta t} \quad \text{(Eq. 1)}$$

where C is the blood lidocaine base concentration at time t and A, α, B, and β are constants. The blood concentration–time data were fitted[5] to Eq. 1 using the NONLIN nonlinear least-squares program (4). Each blood concentration data point was weighted with its squared reciprocal. This weighting factor was selected based on (5):

$$\ln \sigma^2 = \ln a + n \ln \bar{C} \quad \text{(Eq. 2)}$$

where σ^2 is the variance corresponding to the mean drug concentration, $\bar{C}$, for a group of subjects at a given time following injection and a and n are constants. If $n = 1$, an appropriate weighting factor is the reciprocal of the plasma drug concentration; if $n = 2$, the squared reciprocal of the plasma drug concentration may be used. Linear regression of $\ln \sigma^2$ as a function of $\ln \bar{C}$ for the data in this study resulted in n values of 1.39 ($r = 0.970$), 1.59 ($r = 0.923$), and 1.76 ($r = 0.890$) for the 0.5-, 1.0-, and 2.0-mg/kg doses in the pregnant ewes, respectively, and of 1.68 ($r = 0.986$) for the 1.0-mg/kg dose in the nonpregnant ewes. Because the n values approach two, each plasma concentration data point was weighted with its squared reciprocal.

Tables I and II contain estimates of the pharmacokinetic constants (A, α, B, and β) for each subject and values for the criteria used to assess the data fit from each subject to Eq. 1. The criteria were: the coefficient of variation of the estimated pharmacokinetic constants (standard deviation of the estimate/estimated constant), the coefficient of determination [$r^2 = \Sigma(\text{obs}^2 - \Sigma\text{dev}^2)/\Sigma\text{obs}^2$], and the correlation coefficient relating the equation-predicted and observed lidocaine concentrations. The coefficients of determination and correlation coefficients approached

[1] Cardio-Green, Hynson, Wescott and Dunning, Baltimore, Md.
[2] Radiometer BMS3 MK2 blood micro system with PHM71 MK2 acid-base analyzer, London Co., Cleveland, Ohio.
[3] Grass model 7 polygraph with Statham pressure transducers, Quincy, Mass.
[4] Xylocaine, Astra Pharmaceutical Products, Inc., Worcester, Mass.
[5] Using a CDC 6400 computer.

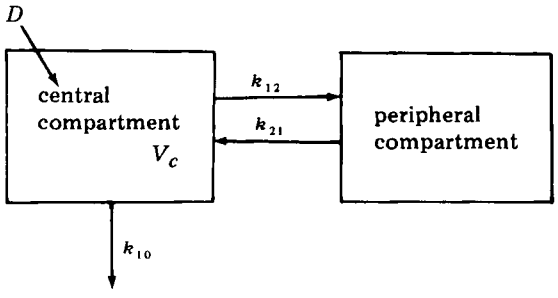
Table III—Pharmacokinetic Parameters in Pregnant Ewes following Intravenous Bolus Lidocaine Hydrochloride Administration (Means ± *SD*)

Dose, mg/kg	*n*	Weight, kg	Cardiac Output, liters/min	$t/2$ (α), min	$t/2$ (β), min	Total Body Clearance, ml/min/kg	V_c, liter/kg	V_{ss} liters/kg	V_B, liters/kg	k_{10}, min^{-1}	k_{12}, min^{-1}	k_{21}, min^{-1}
0.5	5	61.1	7.0	1.6[a]	42.2[b]	38	0.31[a]	1.69[b]	2.24[b]	0.124	0.282[a]	0.063
		±7.3	±1.5	±0.3	±6.5	±8	±0.06	±0.14	±0.24	±0.033	±0.075	±0.020
1.0	4	70.4	7.9[c]	2.0	61.8	42	0.48	2.73	3.63	0.094	0.244	0.052
		±13.4	±2.1	±0.6	±8.6	±13	±0.16	±0.45	±0.80	±0.041	±0.106	±0.028
2.0	5	65.9	6.1	3.2	59.2	34	0.54	2.16	2.83	0.074	0.142	0.044
		±14.3	±1.7	±1.4	±19.4	±7	±0.20	±0.55	±0.69	±0.039	±0.059	±0.012

[a] Statistically significant difference ($p < 0.05$) between the 0.5- and 2.0-mg/kg doses using the Student t test. [b] Statistically significant difference ($p < 0.01$) between the 0.5- and 1.0-mg/kg doses using the Student t test. [c] Three studies.

1.0, indicating that Eq. 1 adequately described the time course of the lidocaine concentration following bolus intravenous administration to pregnant and nonpregnant ewes.

The values for A, α, B, and β were used to calculate (6) pharmacokinetic parameters for the two-compartment open model (Scheme I):



Scheme I

where D represents the intravenous bolus lidocaine hydrochloride dose, V_c is the apparent volume of distribution of the central compartment, k_{12} and k_{21} are the apparent first-order intercompartmental lidocaine distribution rate constants, and k_{10} is the apparent first-order lidocaine elimination rate constant.

RESULTS

The computer-determined estimates of A, α, B, and β for all pregnant animals within each dosage group were averaged and used to generate curves indicating the blood lidocaine concentration change with time following intravenous bolus dosing to pregnant ewes (Fig. 1). The equations in Fig. 1 indicate the average values of A, α, B, and β for each dosage group. Table III shows the means and standard deviations for physiological data and the pharmacokinetic parameters calculated from A, α, B, and β for the three doses in the pregnant ewes.

As the lidocaine dose increased, the slope of the α-phase (distributive phase) of the blood concentration–time curve increased (Fig. 1), as indicated by the increase in the α-phase half-life [$t/2$ (α), Table III]. Although the mean α-phase half-lives for the three doses did not all differ significantly from each other, the α-phase half-life following the 0.5-mg/kg dose differed significantly ($p < 0.05$) from that following the 2.0-mg/kg dose. The curves in Fig. 1 also indicate different slopes for the β-phases (postdistributive phase) of the blood concentration–time curves. These differences are reflected in the β-phase half-lives [$t/2$ (β)] in Table III. The β-phase half-life following the 0.5-mg/kg lidocaine dose differed significantly ($p < 0.01$) from that following the 1.0-mg/kg dose.

The correlation between the total body clearance (calculated by dividing the intravenous bolus dose by the area under the curve for each animal) and the body weight was positive ($r = 0.576$, $p < 0.05$). Therefore, the total body clearance values were normalized for weight prior to averaging within each group (Table III). Cardiac output (mean of the three control period determinations for each study) showed no significant statistical correlation with body weights. However, the correlation between total body clearance and cardiac output was positive ($r = 0.673$, $p < 0.02$, Fig. 2) for all studies performed with the pregnant ewes.

Each of the three volumes of distribution for lidocaine showed statistically significant ($p < 0.05$) positive correlations with body weight and, therefore, were weight normalized for inclusion in Table III. The mean apparent volume of distribution for the central compartment, V_c, following the 0.5-mg/kg dose differed significantly ($p < 0.05$) from that following the 2.0-mg/kg dose. The apparent volume of distribution of

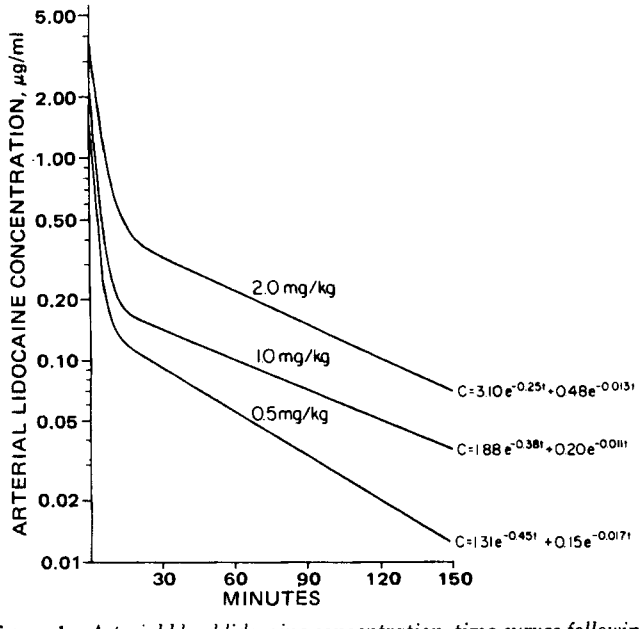


Figure 1—*Arterial blood lidocaine concentration–time curves following intravenous bolus lidocaine hydrochloride administration at three doses to pregnant ewes. The equations indicate the averaged computer estimates for* A, α, B, *and* β *used to generate each curve.*

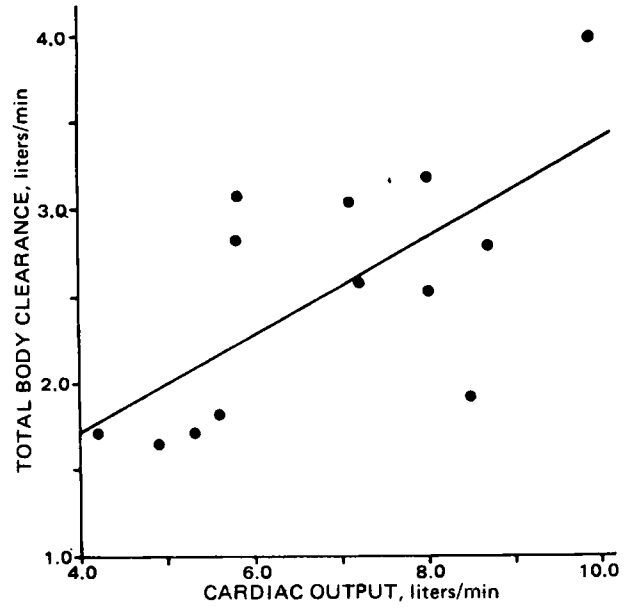


Figure 2—*Correlation between cardiac output and total body clearance of lidocaine following administration of 0.5, 1.0, and 2.0 mg of lidocaine hydrochloride/kg to pregnant sheep.*

Table IV—Individual Pharmacokinetic Parameters in Nonpregnant and Pregnant Ewes following a 1.0-mg/kg Intravenous Bolus Dose of Lidocaine Hydrochloride

Subject	Weight, kg	Cardiac Output, liters/min	$t_{1/2}$ (α), min	$t_{1/2}$ (β), min	Total Body Clearance, ml/min/kg	V_c, liter/kg	V_{ss}, liters/kg	V_B, liters/kg	k_{10}, min^{-1}	k_{12}, min^{-1}	k_{21}, min^{-1}
					Nonpregnant						
1	51.0	—[a]	1.5	34.7	63	0.25	1.60	3.12	0.251	0.204	0.038
2	75.6	6.5	1.1	55.0	23	0.12	1.28	1.83	0.200	0.398	0.039
3	80.2	10.2	3.2	34.8	29	0.34	1.01	1.45	0.085	0.100	0.050
Mean	68.9	8.4	1.9	41.5[b]	38	0.24[c]	1.30[d]	2.13[c]	0.179	0.234	0.042
SD	±15.7	±2.6	±1.1	±11.7	±22	±0.11	±0.30	±0.88	±0.085	±0.151	±0.007
					Pregnant						
1	60.2	5.8	1.7	54.2	47	0.32	2.47	3.68	0.149	0.243	0.036
2	60.9	8.0	2.5	55.5	52	0.71	3.28	4.19	0.074	0.173	0.048
3	71.6	—[a]	1.3	72.7	24	0.43	2.28	2.49	0.055	0.394	0.092
4	89.0	9.9	2.0	64.8	45	0.47	2.91	4.17	0.096	0.165	0.032
Mean	70.4	7.9	2.0	61.8	42	0.48	2.74	3.63	0.094	0.244	0.052
SD	±13.4	±2.1	±0.6	±8.7	±12	±0.16	±0.45	±0.80	±0.041	±0.106	±0.028

[a] Not determined. [b] Statistically significant ($p < 0.05$) difference between nonpregnant and pregnant ewes using the Student t test. [c] Statistically significant ($p < 0.10$) difference between nonpregnant and pregnant ewes using the Student t test. [d] Statistically significant ($p < 0.01$) difference between nonpregnant and pregnant ewes using the Student t test.

lidocaine at steady state, V_{ss}, relates the amount of lidocaine in the body to the blood lidocaine concentration at steady state; the apparent volume of distribution of lidocaine during the postdistributive phase, V_B, relates the amount of lidocaine in the body to the blood lidocaine concentration at any time during the postdistributive, β, phase (6). Mean values for these apparent volumes of distribution also are found in Table III. Both the apparent volume of distribution at steady state and the apparent volume of distribution during the postdistributive phase following the 0.5-mg/kg dose differed significantly ($p < 0.05$) from the corresponding volumes following the 1.0-mg/kg dose.

Table III shows that the lidocaine elimination rate constant, k_{10}, decreased as the dose increased; however, the mean elimination rate constants for each dose did not differ significantly from one another. The rate constants for lidocaine transfer between the central and peripheral compartments also are found in Table III. As the lidocaine dose was increased, both intercompartmental rate constants, k_{12} and k_{21}, decreased.

Figure 3 shows curves generated from averaged computer-determined estimates of A, α, B, and β from pregnant and nonpregnant ewes following 1.0 mg of lidocaine/kg. The Fig. 3 equations indicate the average values of A, α, B, and β. Table IV shows the means and standard deviations of physiological data and pharmacokinetic parameters for 1.0-mg of lidocaine/kg doses in nonpregnant ewes. Comparison of parameters

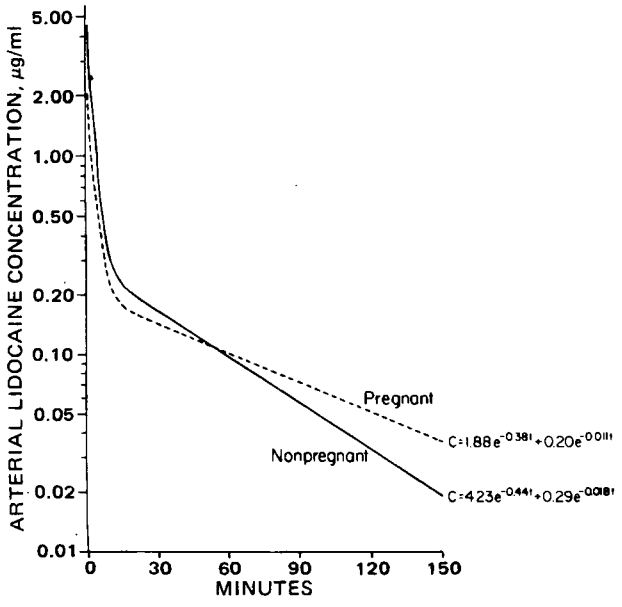


Figure 3—*Arterial blood lidocaine concentration–time curves following intravenous bolus administration of 1.0 mg of lidocaine hydrochloride/kg to pregnant and nonpregnant ewes. The equations indicate the averaged computer estimates for* A, α, B, *and* β *used to generate each curve.*

in the nonpregnant ewe with those in the pregnant ewe (Table IV) shows that the β-phase half-lives differed significantly ($p < 0.05$) between the two groups. Although the volumes of distribution and clearances in this limited sample of nonpregnant ewes did not correlate with weight, the values for these parameters were weight normalized for comparison with the pregnant animal group. The volume of distribution at steady state in the nonpregnant animal differed significantly ($p < 0.01$) from that in the pregnant animal. In addition, the apparent volume of distribution of the central compartment and during the postdistributive phase differed significantly between the two groups but at a lower level of significance ($p < 0.10$).

DISCUSSION

The use of the two-compartment open model (Scheme I) to describe lidocaine disposition in the pregnant ewe implies linearity of lidocaine disposition kinetics as a function of dose. The total body clearance for lidocaine (Table III) remained relatively constant as the intravenous bolus dose was increased from 0.5 to 2.0 mg/kg. Also, the area under the blood lidocaine concentration–time curve, AUC, showed a statistically significant ($r = 0.921$, $p < 0.001$, Fig. 4) linear correlation with the weight-normalized dose, D, according to:

$$AUC = \frac{1}{Cl_T}(D) \qquad \text{(Eq. 3)}$$

where Cl_T represents total body clearance. The regression line passed through the vertical axis near zero (−2.0 min × mg/liter), and the reciprocal of the slope (total body clearance) was 38 ml/min/kg.

Another linearity test in pharmacokinetics involves computer fitting of the blood drug concentration–time data for each animal to an appropriate linear pharmacokinetic model, computing and averaging the pharmacokinetic parameters for each dose, and comparing these averaged parameters among doses. The dose-related trends in the magnitudes of the pharmacokinetic parameters provide strong evidence of nonlinear kinetics (7). Comparisons of averaged pharmacokinetic parameters among different doses in pregnant ewes in this study can be made from the data in Table III. These data showed dose-related increases in the apparent volume of distribution of the central compartment and in the half-life of the α-phase and dose-related decreases in k_{10}, k_{12}, and k_{21} as the dose was increased from 0.5 to 2.0 mg/kg.

The dose-related increase in the apparent volume of distribution of the central compartment could be a result of the hemodynamic effects of lidocaine. Lidocaine is known to increase cardiac output in humans (at arterial lidocaine concentrations of 4–7 μg/ml) and in dogs (8–10), presumably by a central nervous system (CNS) effect evoked through the sympathetic nervous system. Following a bolus lidocaine injection, transient high lidocaine levels in the CNS could produce a brief, dose-related increase in cardiac output. Resulting tissue perfusion increases would promote more rapid tissue uptake of lidocaine and, therefore, an apparent dose-related increase in the volume of distribution of the central compartment. The effect of lidocaine on cardiac output and, hence, on tissue uptake would be brief and would not significantly alter lidocaine elimination; therefore, total body clearance of lidocaine remained un-

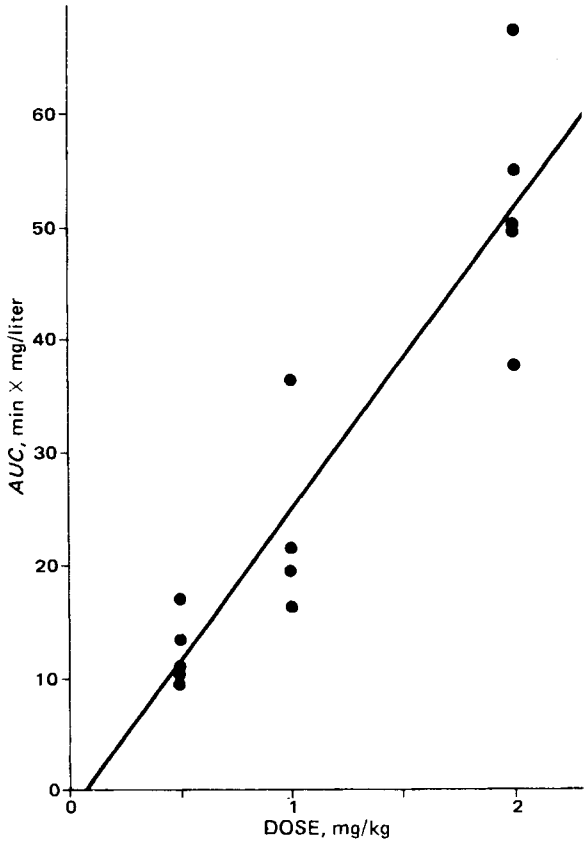


Figure 4—*Correlation between dose and area under the blood lidocaine concentration–time curve following administration of 0.5, 1.0, and 2.0 mg of lidocaine hydrochloride/kg to pregnant sheep.*

changed with dose (Table III). The brevity of the effect of lidocaine on cardiac output is supported by cardiac output measurements at 5 min following bolus lidocaine[6] injection; these cardiac output measurements were not different from the control values shown in Table III.

The dose-related increase in the apparent volume of distribution of the central compartment also is consistent with a probable decrease in the lidocaine fraction bound in plasma as blood lidocaine levels increase. A transient decrease in the lidocaine fraction bound at high blood lidocaine levels following a bolus dose may allow lidocaine to distribute more widely in the body, thus increasing the distribution volume.

The observation of a dose-related increase in the apparent volume of distribution of the central compartment (and a concomitant decrease in k_{10} and k_{12}) in pregnant sheep is consistent with trends reported describing lidocaine disposition kinetics in normal human volunteers (10). In that study, the apparent volume of distribution of the central compartment increased (0.44–0.48 liter/kg), the elimination rate constant k_{10} decreased (0.24–0.22 min^{-1}), and the intercompartmental rate constant k_{12} decreased (0.066–0.041 min^{-1}) as the intravenous bolus dose was increased (50–100 mg). An increase in the apparent volume of distribution of the central compartment in normal humans is consistent with a probable transient increase in cardiac output following a bolus lidocaine dose. In addition, this volume increase is consistent with a decrease in the percent of lidocaine bound in plasma as blood lidocaine levels increase. In humans, plasma lidocaine binding decreases from 75% at 0.4 μg/ml to 58% at 5 μg/ml (11). The mean extrapolated zero-time lidocaine concentrations in normal humans, 1.80 and 2.79 μg/ml following bolus doses of 50 and 100 mg, respectively (10), are in the concentration range where significant plasma binding changes occur. Thus, although pharmacokinetic parameters may not show statistically significant differences as the dose is increased, dose-related trends in pharmacokinetic parameters may offer increased insight into the drug disposition kinetics.

In addition to possible dose-related nonlinearities in the α-phase of lidocaine disposition, Table III shows evidence for dose-related kinetics during the β- or postdistributive phase in pregnant ewes. Following the 0.5-mg/kg dose, the β-phase half-life (42.2 min) differed significantly from that (61.8 min) following the 1.0-mg/kg dose. The β-phase half-life [$t/2(\beta)$] is related to the total body clearance, Cl_T, and to the apparent volume of distribution during the postdistributive phase, V_B, according to:

$$t/2(\beta) = \frac{0.693 V_B}{Cl_T} \qquad \text{(Eq. 4)}$$

Since the total body clearance of lidocaine in the pregnant ewe does not appear to change with dose, Eq. 4 indicates that an increase in the β-phase half-life of lidocaine may be a function of an increase in the apparent volume of distribution. This concept is consistent with the significant change in the volume of distribution during the postdistributive phase (2.24–3.63 liters/kg, Table III) as the lidocaine dose was increased from 0.5 to 1.0 mg/kg. A better indication of dose-related distribution changes is a comparison of the apparent volumes of distribution at steady state following different doses. This parameter is not dependent on elimination processes as is the apparent volume of distribution during the postdistributive phase. The change in the volume of distribution of lidocaine at steady state from 1.69 to 2.73 liters/kg as the dose was increased from 0.5 to 1.0 mg/kg was significant. Thus, the data indicate that dose-related changes in the apparent volume of distribution of lidocaine in the pregnant ewe may influence the biological half-life.

Lidocaine disposition kinetics in the nonpregnant ewe differed from the lidocaine disposition kinetics in the pregnant ewe (Table IV) at a 1.0-mg/kg dose. Figure 3 indicates that the initial lidocaine levels following a 1.0-mg/kg bolus dose were higher in the nonpregnant ewe, but that during the β-phase the blood levels in the nonpregnant ewe decreased more rapidly than in the pregnant ewe. These blood level changes are reflected in the pharmacokinetic parameters in Table IV. The lower apparent volume of distribution of the central compartment in the nonpregnant ewe resulted in higher initial blood lidocaine levels in the nonpregnant animal following a bolus dose based on body weight. In addition, the lower β-phase half-life in the nonpregnant ewe, 41.4 *versus* 61.8 min in the pregnant ewe, reflected the more rapid decline in blood lidocaine levels during the β-phase in the nonpregnant animal following a bolus dose.

Because total body clearances of lidocaine in the nonpregnant and pregnant animals were similar, the greater apparent volumes of distribution (V_c, V_{ss}, and V_B) in the pregnant animals indicated that differences between disposition kinetics in the nonpregnant and pregnant ewes probably were due to drug distribution changes. These changes may be due to differences between lidocaine binding in the plasma, blood, and/or peripheral tissues of the nonpregnant ewe as compared to binding in the corresponding maternal tissues of the pregnant ewe. In addition, fetal lidocaine uptake in the pregnant ewe undoubtedly influences distribution parameters as determined from maternal blood lidocaine levels.

The values for total body clearance of lidocaine in the pregnant sheep (38 ml/min/kg as determined by the method illustrated in Fig. 4) and in the nonpregnant sheep (38 ml/min/kg, Table IV) were considerably greater than the total body clearance reported for lidocaine in humans (9–14 ml/min/kg) (2, 10). In humans, the total body clearance of lidocaine increases as the cardiac output and, therefore, the hepatic blood flow increase (12, 13). In the pregnant sheep, the total body clearance also increased as the cardiac output (Fig. 2) and, presumably, the hepatic blood flow increased. Hepatic blood flow in normal humans is about 21 ml/min/kg (1.5 liters/min in a 70-kg human), and hepatic blood flow in pregnant and nonpregnant ewes is about 65 and 55 ml/min/kg, respectively (14). Thus, the hepatic blood flow in the ewe is sufficient to account for the comparatively higher total body clearance of lidocaine in sheep.

The proportion of the total drug entering the liver to that metabolized by the liver is the extraction ratio. The extraction ratio, E, may be calculated by (15):

$$E = \frac{Cl_T(1-f)}{Q} \qquad \text{(Eq. 5)}$$

where Cl_T is the total body clearance of drug, f is the drug fraction excreted unchanged from the body, and Q is the hepatic blood flow. Use of Eq. 5 for hepatic lidocaine extraction in the pregnant ewe is based on the assumptions that: (*a*) renal excretion is the only nonmetabolic excretory pathway for lidocaine, (*b*) the dose fraction excreted unchanged in the urine [0.014 (16)] in normal adult sheep is the same as that for pregnant ewes, (*c*) lidocaine metabolism occurs only in the liver, and (*d*) hepatic blood flow [65 ml/min/kg (14)] does not change significantly following the lidocaine injection. Cardiac output in the pregnant sheep measured 5 min following injection of 0.5, 1.0, or 2.0 mg of lidocaine/kg[6]

[6] D. C. Bloedow, D. H. Ralston, and J. C. Hargrove, unpublished results.

did not change significantly from control values; presumably, hepatic blood flow behaves similarly. The extraction ratio for lidocaine in the pregnant sheep was 0.57 using Eq. 5. Similar assumptions and calculations for the nonpregnant sheep [Q = 55 ml/min/kg (14)] yielded an extraction ratio of 0.69. These values are similar to the extraction ratio (~0.68) reported for lidocaine in humans (17).

The results of these studies provide evidence for subtle dose-related lidocaine kinetics in pregnant sheep. Further studies on lidocaine binding in ovine plasma and on cardiac output and blood flow distribution during the initial lidocaine distributive phase in sheep are necessary to substantiate the reasons for the apparent nonlinearity of lidocaine disposition.

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False-Positive Alkaloid Reactions

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Abstract □ A variety of nonnitrogenous oxygenated compounds gave false-positive alkaloid reactions with Dragendorff's spray reagent. These compounds reacted positively if the oxygen function and the β-carbon bonded to the oxygen had high electron density. Thus, aldehydes, ketones, lactones, ethers, esters, epoxides, and peroxides with an ethylene bond or free alkyl groups at the β-carbon gave a positive reaction, provided that the availability of electrons at the oxygen and the β-carbon was not altered by electron withdrawal or hydrogen bonding. Carbonyl, ether, and ethylene functions were shown by IR evidence to be involved in coupling. Nitrogen-free, alkaloid-like acetone artifacts were obtained by interaction with fixed alkali and with acids. These compounds were postulated to be α,β-unsaturated aldol condensation products of acetone. Interaction with ammonia in addition yielded nitrogenous alkaloid-like artifacts.

Keyphrases □ Alkaloids—interference by nonnitrogenous alkaloid-like compounds, structural requirements for false-positive reaction □ Acetone artifacts—nonnitrogenous, alkaloid-like compounds, interference with alkaloid detection, structural requirements for false-positive reaction □ Dragendorff's reagent—false-positive alkaloid reaction with nonnitrogenous oxygenated compounds

Of the numerous reagents described for the detection of alkaloids (1), only a few have reliable sensitivity (2). All of these reagents suffer from nonspecificity. Many nitrogenous and nonnitrogenous plant constituents react with several of these reagents similarly to alkaloids (3–8). Some nonnitrogenous compounds react similarly to alkaloids in giving crystalline salts with acids (8).

BACKGROUND

TLC is the most versatile technique for alkaloid detection, separation, monitoring, identification, and quantitation. Dragendorff's spray, in its different modifications, is usually used for visualization of alkaloidal spots on paper and thin-layer chromatograms and in field tests with alkaloid test paper (9–11). Many reports have discussed its sensitivity and specificity (2), false reactions (3, 12), and modifications (13–20). However, many nonnitrogenous plant constituents react with this reagent in a manner typical of alkaloids (3, 12, 21). Such compounds may create difficulty, especially during alkaloid screening without sufficient partition purification steps (4). Complete elimination of these constituents cannot be accomplished through the one partition purification step required in many reported screening procedures (22–26). Moreover, significant amounts of such nonalkaloidal constituents may be detected even after further partition purification (12).

Farnsworth *et al.* (3) determined that any nonnitrogenous compound having conjugated carbonyl or lactone functions would react in a manner typical of alkaloids. The range of compounds that produce a false-positive reaction is greater than is generally realized (27), and compounds such as the common plant sterols and triterpenes are readily detected by Dragendorff's spray.

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