The uptake of isoflurane by the foetal lamb *in utero*: effect on regional blood flow

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Because isoflurane has recently been approved for clinical use in anaesthesia, we have studied the effect of this agent in the foetus using the pregnant ewe as an animal model.

Eight pregnant ewes of 120–125 days gestation were surgically prepared with indwelling catheters and tracheostomy. Prior to anaesthesia, labelled microspheres were injected into the foetal circulation to determine cardiac output and regional blood flows to all organs. The ewes were anaesthetized with a constant inspired concentration of 2.0 per cent isoflurane in oxygen. Blood samples were drawn to construct an uptake curve for both mother and foetus. At 60 and 96 minutes of anaesthesia, microspheres were injected into the foetal circulation to measure changes in organ blood flow from the control period.

Isoflurane crossed the placenta and appeared in the foetal circulation within two minutes. By 96 minutes, maternal and foetal arterial levels were 116.3 ± 9.9 and 99.3 ± 9.1 mEq/L (0.98 vol% and 0.75 vol%). There were no significant changes in foetal blood pressure or pulse rate but foetal pH decreased significantly from 7.39 ± 0.02 to 7.26 ± 0.2 (mean $\pm$ SEM) and base excess decreased from -1.1 ± 1.5 to -6.2 ± 0.7 . Foetal cardiac index decreased from 390.8 ± 26.9 ml·kg⁻¹·min⁻¹ to 292.0 ± 13.8 after 96 minutes of anaesthesia. There were no significant changes in any of the maternal cardiovascular or acid-base parameters.

In the foetal lamb, isoflurane anaesthesia produces foetal acidosis and decreases foetal cardiac index after 96 minutes of anaesthesia.

Key words

ANAESTHETICS, VOLATILE: isoflurane; ANAESTHESIA: obstetric; UTERUS: blood flow.

Isoflurane, a relatively new halogenated inhalation anaesthetic, has received extensive investigation in many clinical trials.^{1–3} The main advantages of this agent are its ease of administration,⁴ its minor degree of metabolism⁵ in the body and its less depressant effect on the cardiovascular system compared to halothane.⁶ This agent does not sensitize the myocardium to catecholamines and, thus, should not produce arrhythmias on this basis.⁷ Isoflurane produces peripheral musculo-skeletal vasodilatation and lowers total peripheral resistance.⁸ It increases heart rate slightly and at 1 MAC levels cardiac output is maintained at awake values.⁹

Because of the above advantages of isoflurane for adults, this agent would be an acceptable alternative to halothane, or enflurane, in the parturient. Using the pregnant ewe as an animal model, Palahniuk and Shnider¹⁰ previously examined the effects of isoflurane on foetal cardiovascular haemodynamics and acid-base status and found that the effects were similar to halothane. Both agents significantly decreased foetal mean arterial blood pressure (MABP) but did not effect oxygenation or acid-base status.

We have used this same animal model but, in addition to the above factors, have measured foetal uptake of isoflurane in order to correlate haemodynamic changes with isoflurane blood levels and have, by using radioactive labelled microspheres, determined the effect of isoflurane on regional blood flows in the foetus.

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Methods

(a) Preparation of the model

Eight pregnant ewes of 125–135 days gestation (term 145–150 days) were surgically prepared as follows: under halothane in oxygen anaesthesia with controlled ventilation, polyvinyl catheters were inserted via a groin incision into the maternal femoral artery and vein. Through a midline abdominal incision the uterus was exposed and a small hysterotomy was made. Through the hysterotomy, foetal limbs were withdrawn for insertion of polyvinyl catheters into the foetal femoral artery and vein and axillary artery. All incisions were closed and the catheters tunnelled subcutaneously to the maternal flank and enclosed in a protective pocket. Each animal was allowed to recover for 24–36 hours prior to the study.

(b) Study period

Prior to each study a tracheostomy was done on each ewe under local anaesthesia. Humidified oxygen was then administered through the tracheostomy at a flow of 10–12 litres. Each study began with a control period of 30 minutes with the ewe standing quietly in an experimental cart to ensure stability of the preparation. Maternal and foetal mean arterial blood pressure (MABP) and pulse rate were recorded continuously via the femoral arterial catheters on a Hewlett-Packard multi-channel recorder (Model 7758C). Maternal and foetal arterial blood gases and pH were measured at 15-minute intervals using a Corning blood gas analyser (Model 165/2). All foetal blood gas samples were drawn from the axillary artery.

In five studies, at the end of the control period, the first injection of labelled microspheres, either cerium¹⁴¹, strontium⁸⁵, or chromium⁵¹ (3M Company, Minneapolis) was made into the foetal femoral vein. A suspension of 0.2 cc containing 1.4×10^6 spheres of 15 ± 0.5 microns in diameter was injected over 15 seconds and flushed with 10 cc of normal saline. Blood was constantly withdrawn from both the axillary and femoral arterial cannulas at a rate of $1.6 \text{ ml} \cdot \text{min}^{-1}$ using a Harvard Pump (Model 940) beginning 30 seconds prior to injection and continuing for 150 seconds following the microsphere injection. The blood samples were weighed and the radioactivity counts were determined by a Searle Analytic Mark III gamma counter (Model No. 6880).¹¹ These served as reference

samples for blood flow to organs above and below the ductus arteriosus.

Isoflurane, at an inspired concentration of 2.0 per cent, was then added to the maternal breathing circuit. After approximately five minutes, ventilation was controlled to maintain maternal blood gases at control values. Two millilitres of arterial blood was withdrawn from the maternal femoral and foetal axillary arteries at 0, 2, 4, 8, 16, 24, 32, 48, 64, and 96 minutes of isoflurane administration. The blood was drawn into heparinized glass syringes and stored at 0°C until the end of the experiment when the samples were analysed by gas-liquid chromatography.¹² During anaesthesia, the ewe received 1000 cc of normal saline intravenously. As the ewe became anaesthetized and ventilation controlled, she was placed in a sitting position.

At 60 and 90 minutes of isoflurane anaesthesia, labelled microspheres were injected into the foetal circulation in the same manner as in the control period, using a different isotope label for each injection.

At the end of each study the animals were killed by an injection of thiopental sodium and saturated KCl and the foetus and placenta removed. The foetus was dissected and each organ system, including the skin and carcass, weighed, ground and ashed. The ashed samples were counted for radioactivity and these counts were compared to the reference samples to calculate the blood flow in each organ system. The total of all organ blood flows was considered to be the cardiac output of the foetus. This slightly underestimates true biventricular cardiac output because we did not have a reference sample for the lung, so did not calculate pulmonary blood flow.¹¹ To correct for differences in foetal size, which is considerable during the last 20 days gestation in the sheep, the cardiac output was converted into a foetal cardiac index by dividing by foetal weight.

Analysis of results for maternal and foetal mean arterial blood pressure, pulse rate, blood gases and pH changes was by analysis of variance. Maternal and foetal isoflurane levels at each time period were compared by paired "t" test. Foetal regional blood flows and cardiac index changes for control, 60 and 90 minutes of isoflurane anaesthesia were tested for significance by analysis of variance. "P" < 0.05 was considered significant.

TABLE I Maternal and foetal mean arterial pressure and pulse rate during administration of 2.0% isoflurane to the pregnant ewe

		Time (minutes)							
	Control	2	8	16	24	32	48	64	96
<i>Maternal</i>									
MABP	96.0	96.0	83.5	83.0	84.0	86.3	89.5	96.0	97.8
(torr)	± 1.1	± 4.9	± 7.46	± 6.1	± 5.3	± 6.9	± 7.7	± 8.4	± 9.1
Pulse rate	102.6	112.5	102.6	111.0	99.4	93.1	98.1	107.5	106.2
(beats/min)	± 3.2	± 6.8	± 7.4	± 11.2	± 3.8	± 4.9	± 4.8	± 6.5	± 5.6
<i>Foetal</i>									
MABP	63.6	58.0	51.0	50.0	50.8	50.5	50.0	49.2	50.2
(torr)	± 3.4	± 3.2	± 3.5	± 3.1	± 3.9	± 3.9	± 3.7	± 3.8	± 4.3
Pulse rate	164.0	169.0	165.0	166.0	160.0	158.0	156.0	156.0	146.0
(beats/min)	± 8.7	± 9.1	± 9.6	± 13.5	± 13.5	± 7.9	± 7.5	± 8.4	± 8.4

N = 8, Mean ± S.E.

TABLE II Maternal and foetal blood gases and pH during administration of isoflurane to the pregnant ewe (2.0% inspired concentration)

		Time (minutes)						
	Control	8	16	24	32	48	64	96
<i>Maternal</i>								
pH (units)	7.50 ± 0.02	7.44 ± 0.03	7.48 ± 0.03	7.51 ± 0.03	7.50 ± 0.02	7.50 ± 0.02	7.49 ± 0.02	7.47 ± 0.03
H ⁺ nanomoles/L	31.6 ± 1.4	36.3 ± 2.5	33.1 ± 1.5	30.0 ± 2.2	31.6 ± 1.4	31.6 ± 1.4	32.4 ± 1.5	33.9 ± 1.5
PCO ₂	29.0 ± 1.4	33.0 ± 2.1	30.0 ± 2.2	30.0 ± 1.1	28.0 ± 0.7	30.0 ± 1.2	29.0 ± 1.5	34.0 ± 2.6
PO ₂	185.0 ± 4.1	351.0* ± 30.4	353.0* ± 27.8	363.0* ± 29.8	363.0* ± 24.8	361.0* ± 31.6	350.0* ± 30.2	322.0* ± 20.8
B.E.	2.4 ± 0.8	-0.6 ± 1.0	1.4 ± 0.8	2.4 ± 1.3	2.0 ± 1.5	2.1 ± 1.0	0.9 ± 0.8	2.4 ± 1.1
<i>Foetal</i>								
pH (units)	7.39 ± 0.02	7.35 ± 0.02	7.38 ± 0.02	7.37 ± 0.02	7.35 ± 0.01	7.34* ± 0.01	7.33* ± 0.02	7.26* ± 0.02
H ⁺ nanomoles/L	40.7 ± 1.8	44.7 ± 2.1	41.7 ± 1.9	40.7 ± 1.9	44.7 ± 2.1	45.7* ± 1.0	46.8* ± 2.1	54.9* ± 2.2
PCO ₂	37.0 ± 1.8	44.0 ± 2.4	42.0 ± 2.8	40.0 ± 2.2	39.0 ± 2.2	40.0 ± 1.7	41.0 ± 2.2	44.0 ± 2.9
PO ₂	20.0 ± 1.2	24.0 ± 1.6	26.0 ± 1.3	25.0 ± 1.2	24.0 ± 1.2	24.0 ± 1.2	24.0 ± 1.3	25.0 ± 1.5
B.E.	-1.1 ± 1.5	-0.8 ± 0.9	+0.1 ± 1.8	-2.1 ± 0.6	-2.4 ± 0.6	-2.6 ± 0.7	-3.2* ± 0.9	-6.2* ± 0.7

*p < 0.05.

N = 8, Mean ± S.E.

Results

During the administration of isoflurane to the pregnant ewe, maternal MABP tended to decline but no values were significantly different from control. There were no significant changes in pulse

rate (Table I). Maternal PCO₂, pH and BE did not vary significantly from control values. Maternal PO₂ increased significantly after eight minutes of isoflurane anaesthesia with the institution of controlled ventilation (Table II).

TABLE III The effect of isoflurane on foetal cardiac index* and regional blood in the normal foetal lamb *in utero*

Organ blood flow $\text{ml} \cdot \text{gm}^{-1} \cdot \text{min}^{-1}$	Isoflurane		
	Control	60 min	90 min
Brain	1.12 ± 0.17	1.06 ± 0.20	1.25 ± 0.11
Myocardium	1.12 ± 0.23	0.88 ± 0.13	0.95 ± 0.11
Splanchnic bed	1.64 ± 0.35	$1.46 \pm 0.31^\dagger$	1.52 ± 0.51
Musculo-skeletal	0.34 ± 0.03	0.28 ± 0.03	0.29 ± 0.03
Skin	0.15 ± 0.02	0.11 ± 0.02	0.11 ± 0.02
Placenta	1.13 ± 0.11	$0.86 \pm 0.09^\dagger$	$0.83 \pm 0.11^\dagger$
Cardiac index* $\text{ml} \cdot \text{kg}^{-1} \cdot \text{min}^{-1}$	390.8 ± 26.9	$286.0 \pm 21.7^\dagger$	$292.0 \pm 13.8^\dagger$
Foetal isoflurane arterial blood level volume %	0	0.73 ± 0.05	0.75 ± 0.08

*Foetal cardiac index = total cardiac output divided by foetal weight.

 $^\dagger p < 0.05$ significantly different from control.N = 5, Mean $\pm$ S.E.

TABLE IV Maternal and foetal isoflurane levels during administration of isoflurane to the pregnant ewe at an inspired concentration of 2.0%

	Time (minutes)									
	0	2	4	8	16	24	32	48	64	96
Maternal arterial concentration $\text{mg} \cdot \text{L}^{-1}$	0	28.7 ± 3.4	54.5 ± 9.5	72.6 ± 9.7	86.9 ± 11.7	97.1 ± 11.1	94.5 ± 9.9	105.6 ± 7.7	117.8 ± 12.2	116.3 ± 9.9
Foetal arterial concentration $\text{mg} \cdot \text{L}^{-1}$	0	15.2* ± 4.3	23.5* ± 5.5	34.5* ± 5.2	53.0* ± 6.8	58.8* ± 5.7	66.7* ± 8.2	77.5* ± 7.9	85.7* ± 5.8	99.3* ± 9.1

* $p < 0.05$ foetal levels significantly lower than maternal.(N = 8, Mean $\pm$ S.E.).

Foetal MABP declined from a control of 63.4 ± 3.4 torr to 51.0 ± 3.5 torr after eight minutes of isoflurane anaesthesia and remained at this level for the duration of the experiment. This did not represent a significant difference from control values, however. Foetal heart rate was unchanged from control (Table I).

Foetal blood gases and pH remained stable until 48 minutes of isoflurane anaesthesia. From that point onward there was a significant fall in foetal pH and base excess. At 96 minutes of anaesthesia, foetal pH had declined from a control value of 7.39 ± 0.02 (mean $\pm$ SEM) to 7.26 ± 0.02 . Base excess had decreased from -1.1 ± 1.5 to -6.2 ± 0.7 .

Analysis of the microsphere data revealed a

significant decrease in foetal cardiac index at both 60 and 96 minutes of isoflurane anaesthesia (Table III). This was accompanied by a significant fall in blood flow to the placenta at both 60 and 96 minutes and a significant decrease in splanchnic blood flow at 60 minutes, which returned to near control values by 96 minutes.

The uptake of isoflurane by the foetus is shown in Table IV in mEq/l and in the Figure converted into volume per cent. Isoflurane crosses the placenta rapidly and appears in the foetal circulation within two minutes. Foetal arterial levels remain significantly below maternal arterial levels throughout the 96 minutes of anaesthesia. The uptake curve for the foetus parallels the maternal curve.

FIGURE Maternal and foetal arterial isoflurane levels during administration of isoflurane at an inspired concentration of 2.0% for 96 minutes. (N = 8 Mean $\pm$ S.E.)

Discussion

Isoflurane uptake by the foetus is similar to that of halothane.¹³ Isoflurane crosses the placenta rapidly but foetal arterial levels rise slowly and after 90 minutes are still significantly less than maternal arterial levels. This may demonstrate the relative inefficiency of the placenta compared to the lung. Placental shunting in both the maternal and foetal circulations has been previously demonstrated¹⁴ and may account for the slower rise in foetal blood levels of isoflurane. Uptake of isoflurane by the foetal liver may also occur and, in part, account for the lower foetal arterial levels. The microsphere data demonstrate a significant fall in foetal placental blood flow. This would also impede uptake of isoflurane.

In our study of the normal foetal lamb, the MABP declined 20 per cent after eight minutes of isoflurane anaesthesia (P = NS, analysis of variance). This is in contrast to our results with halothane (a significant fall of 33 per cent)¹³ and, also, the results obtained by Palahniuk and Shnider¹⁰ who found a progressive decrease in foetal MABP at 1 and 2 MAC isoflurane. In our study, by 96 minutes, the maternal level of isoflurane was 1 MAC (0.98 vol%).¹⁰ We did not demonstrate an increase in musculo-skeletal blood flow in the foetus either,

as had been found in human studies with isoflurane. In the foetal lamb, isoflurane did appear to depress myocardial function as the foetal cardiac index declined significantly after 60 minutes of anaesthesia. The deterioration of foetal pH and base excess by 48 minutes of anaesthesia indirectly suggests that cardiac depression may have been occurring even earlier. As heart rate did not change during the period of anaesthesia, a decline in stroke volume, and thus contractility, likely accounts for the fall in cardiac index.

We also found that foetal blood flow to the placenta decreased significantly with isoflurane anaesthesia. This is in contrast to lambs anaesthetized with halothane, in which placental blood flow did not decline significantly.¹⁵ The decreased flow may represent shunting of blood away from the placenta, but this is unlikely because blood flow to other organs did not increase. It is probably a reflection of decreasing cardiac output. Splanchnic blood flow also decreased initially but then increased by 90 minutes to control values.

The effects of isoflurane on foetal status is one of progressive foetal acidosis, although significant acidosis and myocardial depression did not occur until one hour of anaesthesia. This is a much longer exposure than one would have, clinically, at Caesarean section or vaginal delivery. The normal foetus would likely tolerate short exposures to isoflurane, but the depressant effect in the stressed foetus might occur more quickly.

In view of the fact that halothane, in contrast to isoflurane, does not produce acidosis or a fall in foetal cardiac index – even after 90 minutes – there seems little reason at present to suggest that isoflurane be used instead of halothane for general anaesthesia in obstetrics.

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Résumé

L'approbation récente de l'isoflurane pour usage clinique en anesthésie nécessitait une étude de ses effets sur le fœtus; elle fut réalisée sur la brebis gravide.

Des sondes à demeure ont été installées par manœuvres chirurgicales sur huit brebis gravides de 120 à 125 jours qui furent aussi trachéotomisées. Avant l'anesthésie, des microsphères marquées ont été injectées dans la circulation fœtale pour mesurer le débit cardiaque et les débits sanguins régionaux. Les brebis furent anesthésiées par inhalation d'une concentration inspirée constante de 2 pour cent d'isoflurane dans l'oxygène. Des échantillons furent prélevés pour tracer une courbe de capture tant pour la mère que pour le fœtus. Après 60 et 96 minutes d'anesthésie, des microsphères furent injectées dans la circulation fœtale pour mesurer les changements de débits régionaux comparatifs à la période de contrôle.

L'isoflurane a traversé le placenta et est apparu dans le sang fœtal en moins de deux minutes. À la 96ième minute, les niveaux artériels maternels et fœtaux étaient respectivement de 116.3 ± 9.9 et de 99.3 ± 9.1 (0.98 vol pour cent et 0.75 vol. pour cent). Il n'y a pas eu de changements significatifs de la pression sanguine et du pouls fœtaux mais le pH a baissé de façon significative de 7.39 ± 0.02 à 7.26 ± 0.2 et l'excès de base est passé de -1.1 ± 1.5 à -6.2 ± 0.7 . L'index cardiaque fœtal a diminué de $390.8 \pm 26.9 \text{ ml} \cdot \text{kg}^{-1} \cdot \text{min}^{-1}$ à 292.0 ± 13.8 après 96 minutes d'anesthésie. On n'a pas trouvé de changements significatifs dans les paramètres maternels cardiovasculaires et acido-basiques.

L'anesthésie à l'isoflurane produit chez le fœtus de la brebis de l'acidose et diminue l'index cardiaque après 96 minutes d'anesthésie.