**P**$_{O_2}$, **P**$_{CO_2}$ AND pH LEVELS IN THE UMBILICAL AND UTERINE BLOOD OF THE MARE AND EWE

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**SUMMARY**

1. Foetal and maternal blood gas tensions and pH levels have been investigated in the mare and the ewe during late pregnancy under a number of experimental conditions.

2. Observations were made on anaesthetized animals with the foetus *in utero*. Simultaneous blood samples were withdrawn from a maternal artery and uterine vein, and from the two umbilical vessels, through catheters inserted at the beginning of each experiment.

3. At normal maternal arterial **P**$_{O_2}$ (80–110 mm Hg) the umbilical venous **P**$_{O_2}$ of the foal was very high (49 mm Hg) and the **P**$_{O_2}$ in the umbilical artery (33 mm Hg) was similar to that in the umbilical vein of the lamb (34 mm Hg).

4. The **P**$_{O_2}$ difference between the umbilical and uterine venous blood was 17 mm Hg in the ewe but only 4 mm Hg in the mare. The corresponding **P**$_{CO_2}$ gradients were about one quarter of those for oxygen in both species.

5. When the maternal arterial **P**$_{O_2}$ was raised above 100 mm Hg both uterine venous and umbilical **P**$_{O_2}$ increased in the mare, and the gradient between the uterine vein and umbilical vein was reversed, whereas little change occurred in the corresponding vessels of the ewe.

6. Alterations in maternal arterial **P**$_{CO_2}$ were associated with concomitant changes in the blood in the other three vessels but in both species the **P**$_{CO_2}$ difference between uterine and umbilical venous blood appeared to remain constant.

7. Foetal blood pH levels followed those of the mother if the changes in the maternal blood were of respiratory origin. The effects of prolonged changes in maternal pH on foetal levels were investigated in the ewe. Foetal pH remained unchanged during maternal alkalaemia (pH 7.7) induced by Na$_2$CO$_3$ infusions, but increased during a similar rise in maternal pH induced by hyperventilation.
8. The present observations have been compared with findings on the conscious animal and possible explanations for the differences between the two species are discussed.

**INTRODUCTION**

The mechanisms responsible for the placental exchange of blood gases have received much attention in recent years and the subject has been extensively reviewed by Bartels & Metcalfe (1965), Metcalfe, Bartels & Moll (1967) and Dawes (1968). There is, however, a paucity of data for nearly all species other than the sheep, and even here work has been confined mainly to the problem of oxygen transfer across the placenta.

The majority of evidence indicates in the sheep that a large and stable $P_{O_2}$ gradient exists between the uterine vein and the umbilical vessels (Barron, 1951; Parker & Purves, 1967). Furthermore, changes in maternal arterial $P_{O_2}$ have little effect on foetal or uterine venous $P_{O_2}$. Thus foetal hypoxia occurs only after a marked reduction in the maternal arterial blood $P_{O_2}$ and attempts to raise the foetal $P_{O_2}$ have met with little success despite the administration of pure oxygen to the mother (Comline, Silver & Silver, 1965; Kirschbaum, Lucas, De Haven & Assali, 1967; Rivard, Motoyama, Acheson, Cook & Reynolds, 1967; Parker & Purves, 1967). The $P_{O_2}$ gradient across the placenta is still present even when the foetus is supplied with oxygen by other means (Campbell, Dawes, Fishman, Hyman & James, 1966).

In the human the exchange of oxygen across the placenta has also been studied but, for obvious reasons, the majority of the observations have been made at, or immediately before, delivery (Wulf, 1964; Newman, McKinnon, Philips, Paterson & Wood, 1967; Althabe, Schwarcz, Pose, Escarcena & Caldeyro-Barcia, 1967; Walker, Philips, Powe & Wood, 1968). This may account for the conflicting reports on the effects of oxygen administration to the mother on the levels in the foetal blood (Newman et al. 1967).

We have now examined the placental exchange of blood gases in the mare in which the diffuse epitheliochorial placenta is very different from the cotyledonary type of the ruminant; a comparable series of observations have also been made on the pregnant ewe under similar conditions. A preliminary report of some of these findings has already been published (Comline & Silver, 1968).

**METHODS**

**Animals**

Twenty Welsh ewes of known gestational age and eleven small pony mares of approximate gestational age (± 3 weeks) were used for this investigation. Five ewes were used at 34–60 days gestation and all other experiments on both species were
carried out during the last month of pregnancy (duration of gestation; ewe ~ 147 days; mare ~ 340 days). All food but not water was withdrawn from the animals 12–24 hr before the beginning of experiments.

Preparation

**Ewes.** In seventeen animals anaesthesia was induced and maintained by the i.v. injection of sodium pentobarbitone (May & Baker; 6 g/100 ml. in 0·9 % (w/v) NaCl). Closed-circuit halothane (Flurothane, ICI) anaesthesia was used initially in the three other animals; it was administered from a Boyle’s apparatus, which contained a CO₂ absorption canister, in a mixture of O₂ and N₂ adjusted to give normal maternal arterial $P_{O_2}$ levels. In all experiments ventilation was spontaneous unless otherwise stated.

The uterus was exposed by a mid line abdominal incision and vinyl catheters, filled with 0·9 % (w/v) NaCl containing 1000 i.u. heparin/ml. (heparin-saline) were threaded into tributaries of the uterine vein and umbilical vessels according to the technique of Meschia, Cotter, Breathnach & Barron (1965). The position of each catheter was examined at the end of the experiment. In a few animals it was found that a catheter had passed through the main vessel and back into a small branch or had doubled back on itself; the results from these were discarded. Maternal and foetal blood pressures were recorded throughout the experiments and a $P_{O_2}$ electrode (Silver, 1963) placed in a maternal femoral arterio-venous loop (Comline et al. 1965) enabled maternal arterial $P_{O_2}$ to be monitored continuously. When necessary, supplementary $O_2$ was administered to the ewes as a slow stream (1–1·1/min) into a side-arm of the tracheal cannula to maintain a constant resting maternal arterial $P_{O_2}$ (over-all range 80–110 mm Hg).

**Mares.** Four mares were anaesthetized with sodium pentobarbitone (12 g/100 ml. in 0·9 % (w/v) NaCl) and three with chloralose (initial dose up to 70 mg/kg). All these animals respired spontaneously through a tracheal cannula. In addition, samples were taken from four animals under closed-circuit halothane anaesthesia with positive pressure ventilation, during the insertion of catheters under sterile conditions. These four animals were not used for the experimental alteration of blood gases.

The uterus was exposed by a paramedial incision on the left side. A uterine venous catheter was inserted first by passing a length of 15–20 in. vinyl tubing (0·06 in. o.d.) into one of the tributaries. A small incision through the endometrium and placental layers in one of the horns of the uterus usually revealed convenient small umbilical vessels. Vinyl catheters similar to that in the uterine vein were inserted into an umbilical artery and vein, and threaded 10–15 in. towards the main vessels of the umbilical cord. The catheters were tested for withdrawal of blood samples, flushed with heparin-saline and the uterine incision was then closed. In the acute experiments maternal arterial blood samples were taken through a catheter in the facial artery and maternal arterial $P_{O_2}$ was maintained at 80–110 mm Hg (over-all range) by the administration of supplementary oxygen through the tracheal cannula. Maternal and foetal blood pressures were recorded throughout these experiments and at the end the positions of the catheters in the uterine and umbilical vessels were checked as in the ewes.

Alterations in maternal blood gas tensions and pH

$P_{O_2}$. Maternal arterial $P_{O_2}$ was altered by allowing the animals to breathe O₂- or N₂-enriched air for 10–20 min at any given level of administration. In the ewes all changes in maternal arterial $P_{O_2}$ were continuously recorded by the $P_{O_2}$ electrode; in the mares, the maternal arterial $P_{O_2}$ was checked in frequent individual samples. In
both species samples from the umbilical and uterine vessels were withdrawn simultaneously at 5–10 min intervals to ensure that the final measurements were made when equilibrium conditions had been reached in both foetal and maternal circulations.

$P_{CO_2}$. Maternal arterial $P_{CO_2}$ was altered experimentally only in the ewes: hypercapnia was induced (i) by the addition of 5, 7 or 10% CO$_2$ to the inspired air with an appropriate reduction in % O$_2$ to compensate for the hyperventilation which ensued, or (ii) by the use of closed-circuit halothane anaesthesia (see Comline & Silver, 1970). The latter procedure enabled a high and stable arterial $P_{CO_2}$ to be maintained for long periods, even in the presence of a CO$_2$ absorption canister. Hypocapnia was produced by hyperventilation with a Starling–Ideal respiratory pump.

Equilibrium conditions were readily attained under halothane anaesthesia and during hyperventilation under sodium pentobarbitone; these regimes were continued for 15 min–3 hr and simultaneous sets of samples were taken at frequent intervals initially and then at 30 min intervals. Stable conditions were more difficult to establish at high levels of CO$_2$ intake and during hyperventilation under halothane. However, maternal arterial $P_{CO_2}$ was analysed at 2–4 min intervals and the conditions were kept as constant as possible for 5–10 min before the final set of simultaneous samples was withdrawn.

$\text{pH}$. Maternal arterial $\text{pH}$ was altered by the continuous i.v. infusion of 5% (w/v) Na$_2$CO$_3$ in distilled water at rates between 3 and 8 ml./min by means of a Harvard constant infusion pump. Maternal arterial $\text{pH}$ was maintained at 7.7–7.8 under this regime for 2–3 hr. Simultaneous sets of blood samples were taken at 30 min intervals.

**Samples and measurements**

Between twelve and fifteen sets of samples were taken in acute experiments over a period of 2–3 hr after the completion of all operative procedures, i.e. observations were started 45–90 min and 2–3 hr after the induction of anaesthesia in the ewes and mares respectively. In addition, frequent maternal arterial samples were taken at 10–15 min intervals to check $P_{CO_2}$, $\text{pH}$, and in the mare, $P_{O_2}$ levels. All samples were taken simultaneously into 2 ml. syringes, the dead space of which was filled with heparin-saline. Blood (0.5–1.0 ml.) was taken from each foetal vessel in the lambs and in some experiments samples from the umbilical artery were omitted; all other samples were 2–0 ml. The majority of samples were analysed immediately, others were stored at 4°C for ½–1 hr when necessary. Owing to its rapid sedimentation rate, maternal equine blood was drawn into syringes which contained small brass washers to ensure thorough mixing.

$\text{pH}$, $P_{CO_2}$ and $P_{O_2}$ measurements were all made at 38.5°C with standard Radiometer (Copenhagen) equipment. Further details are given elsewhere (Comline & Silver, 1970).

**RESULTS**

**Resting $P_{O_2}$, $P_{CO_2}$ and $\text{pH}$ in uterine and umbilical vessels**

Table 1 summarizes the data obtained from seven ewes and eleven mares: all values were obtained when maternal arterial $P_{O_2}$ remained between 80 and 110 mm Hg. In the sheep maternal arterial $P_{CO_2}$ and $\text{pH}$ varied little under resting conditions and even after 2–3 hr under sodium pentobarbitone anaesthesia $P_{CO_2}$ did not rise above 40 mm Hg and $\text{pH}$ remained above 7.45. Similar conditions were more difficult to maintain in the mares and in the four experiments with sodium pentobarbitone
Table 1. Mean resting $P_{O_2}$, $P_{CO_2}$ and pH levels (±s.e.) in foetal and maternal blood of seven ewes and four mares anaesthetized with sodium pentobarbitone (NP) and seven mares anaesthetized with chloralose or halothane (C/H)

<table>
<thead>
<tr>
<th></th>
<th>Blood samples from</th>
<th>$P_{O_2}$ (mm Hg)</th>
<th>$P_{CO_2}$ (mm Hg)</th>
<th>pH</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Ewe (NP)</td>
<td>Mare (C/H)</td>
<td>Mare (NP)</td>
<td>Ewe (NP)</td>
</tr>
<tr>
<td>Artery</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Maternal</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Uterine vein</td>
<td>(n = 11)</td>
<td>(n = 12)</td>
<td>(n = 7)</td>
<td>(n = 11)</td>
</tr>
<tr>
<td>Vein</td>
<td>(n = 11)</td>
<td>(n = 12)</td>
<td>(n = 7)</td>
<td>(n = 11)</td>
</tr>
<tr>
<td>Foetal umbilical</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Artery*</td>
<td>(n = 4)</td>
<td>(n = 12)</td>
<td>(n = 5)</td>
<td>(n = 4)</td>
</tr>
</tbody>
</table>

Number of observations (n) given in parentheses (1-2 observations per animal).
* Umbilical artery not catheterized in four of the ewes, and one mare (NP).
anaesthesia, respiratory acidosis invariably developed before the operative procedures had been completed, although maternal arterial $P_{O_2}$ was within normal limits in these animals. The remaining seven mares were anaesthetized either with chloralose (three animals) or with halothane administered under positive pressure ventilation (four animals); under these conditions maternal gas tensions and pH levels were very similar to those found in the ewes (Table 1).

At comparable maternal blood gas and pH levels, there was a most striking difference between the foetal $P_{O_2}$ levels in the two species (Table 1), for the mean $P_{O_2}$ in the umbilical venous blood of the foal was 49 mm Hg and that in the lamb only 34 mm Hg. Furthermore, in the foal, umbilical venous $P_{O_2}$ was very close to that in the uterine vein so that the mean $P_{O_2}$ gradient between the two vessels was small (3·7 ± 1·3 mm Hg), whereas the corresponding gradient in the ewe (16·9 ± 0·9 mm Hg) was 4-5 times greater. The $P_{O_2}$ in the umbilical vein of the foal remained close to that in the uterine vein whether maternal $P_{CO_2}$ was normal or high, but the absolute levels in both vessels were 9-10 mm Hg higher when maternal $P_{CO_2}$ was 25-30 mm Hg above normal. This presumably indicates a Bohr effect on both maternal and foetal blood.

There appeared to be virtually no $P_{CO_2}$ gradient between the umbilical and uterine venous blood in the mare whereas a consistent difference of about 4 mm Hg was found between the two vessels in the ewe. In fact, the fourfold difference between the two species for $P_{CO_2}$ was of the same order as the difference between their respective $P_{O_2}$ gradients across the placenta.

The arterio-venous $P_{O_2}$ difference across the umbilical vessels was about 12 mm Hg in the lamb and 16 mm Hg in the foal. In spite of the larger difference in the foal, the $O_2$ tension of the blood entering the equine placenta from the foetus (32·8 mm Hg) was similar to that leaving the placenta of the foetal lamb after exchange (34·4 mm Hg).

Comparison with data from conscious animals

Although the figures summarized in Table 1 were obtained with minimum disturbance of the foetus, they represented normal resting levels only within the conditions of the experiments. It was therefore important to compare these data with values obtained in the conscious pregnant animal with catheters implanted in the same way into the uterine and umbilical vessels. Some of the results obtained in a large series of conscious ewes (Comline & Silver, 1970) are summarized in Table 2, together with some preliminary observations on blood gas levels and pH in the conscious pregnant mare with indwelling catheters in umbilical and uterine vessels (R. S. Comline, L. W. Hall, M. Silver & R. G. Walker, unpublished observations). Differences between the foetal $P_{O_2}$ levels in conscious animals of
Table 2. Mean resting $P_{O_2}$, $P_{CO_2}$ and pH levels (+ s.e.) in foetal and maternal blood of conscious ewes* and mares†

<table>
<thead>
<tr>
<th>Blood samples from</th>
<th>Ewe ($P_{O_2}$ mm Hg)</th>
<th>Mare ($P_{O_2}$ mm Hg)</th>
<th>Ewe ($P_{CO_2}$ mm Hg)</th>
<th>Mare ($P_{CO_2}$ mm Hg)</th>
<th>Ewe</th>
<th>Mare</th>
</tr>
</thead>
<tbody>
<tr>
<td>Artery</td>
<td>98.9 ± 1.1</td>
<td>95.3 ± 2.3</td>
<td>33.6 ± 0.4</td>
<td>35.3 ± 1.0</td>
<td>7.478 ± 0.007</td>
<td>7.427 ± 0.008</td>
</tr>
<tr>
<td>(n = 15)</td>
<td>(n = 14)</td>
<td>(n = 15)</td>
<td>(n = 14)</td>
<td></td>
<td>(n = 15)</td>
<td>(n = 14)</td>
</tr>
<tr>
<td>Uterine vein</td>
<td>53.1 ± 1.1</td>
<td>50.2 ± 1.2</td>
<td>37.9 ± 1.1</td>
<td>38.4 ± 1.1</td>
<td>7.440 ± 0.009</td>
<td>7.408 ± 0.004</td>
</tr>
<tr>
<td>(n = 9)</td>
<td>(n = 12)</td>
<td>(n = 9)</td>
<td>(n = 11)</td>
<td></td>
<td>(n = 9)</td>
<td>(n = 11)</td>
</tr>
<tr>
<td>Vein</td>
<td>34.8 ± 1.3</td>
<td>48.5 ± 1.3</td>
<td>41.5 ± 0.8</td>
<td>39.3 ± 1.0</td>
<td>7.399 ± 0.006</td>
<td>7.404 ± 0.007</td>
</tr>
<tr>
<td>(n = 15)</td>
<td>(n = 11)</td>
<td>(n = 15)</td>
<td>(n = 10)</td>
<td></td>
<td>(n = 15)</td>
<td>(n = 10)</td>
</tr>
<tr>
<td>Foetal umbilical</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>7.346 ± 0.009</td>
<td>7.346 ± 0.009</td>
</tr>
<tr>
<td>Artery</td>
<td>22.8 ± 1.2</td>
<td>33.4 ± 0.9</td>
<td>45.7 ± 1.8</td>
<td>46.2 ± 1.5</td>
<td>(n = 7)</td>
<td>(n = 8)</td>
</tr>
<tr>
<td>(n = 7)</td>
<td>(n = 8)</td>
<td>(n = 7)</td>
<td>(n = 8)</td>
<td></td>
<td>(n = 7)</td>
<td>(n = 8)</td>
</tr>
</tbody>
</table>

* Seven ewes with either or both foetal catheters patent for 4–17 days (data from Comline & Silver, 1970); one to three observations per ewe.

† Five mares with either or both foetal catheters patent for 2–4 days (R. S. Comline, L. W. Hall, M. Silver & R. G. Walker, unpublished observations); two to four observations per mare.
the two species were of the same magnitude as those found in anaesthetized preparations.

In each species good agreement was found between values in conscious and anaesthetized animals. In the mare, apart from the results obtained under sodium pentobarbitone anaesthesia, the values for $P_{O_2}$ and $P_{CO_2}$ in

![Graph showing changes in $P_{O_2}$ in the umbilical artery (●), umbilical vein (○) and uterine vein (●) at different levels of maternal arterial $P_{O_2}$ (●). Ewe and mare (a), sodium pentobarbitone anaesthesia; mare (b), chloralose anaesthesia. Note: each point shows the $P_{O_2}$ attained after a period of 10–20 min at a given level of maternal $O_2$ intake.]

maternal and foetal blood during anaesthesia did not differ significantly from those found in the conscious animal. The slight differences in pH between conscious and anaesthetized mares were statistically significant and probably reflect the metabolic acidosis which develops in the horse
during anaesthesia when $P_{CO_2}$ remains low or is controlled at a low level. In the ewes, only the difference in maternal arterial $P_{CO_2}$ between conscious and anaesthetized animals was significant ($P < 0.05$).

**Effects of changes in maternal arterial $P_{O_2}$ on umbilical and uterine $P_{O_2}$ levels**

Alterations in maternal $P_{O_2}$ without significant changes in $P_{CO_2}$ or pH were produced by the administration of gas mixtures. The blood gas tensions and pH in the umbilical vessels, the uterine vein and maternal artery were followed intermittently and the final determinations, once a steady state had been reached, were made after 10–20 min on a given regime. Fig. 1 shows the $P_{O_2}$ changes observed in three such experiments: each set of four points indicates the $P_{O_2}$ in foetal and maternal blood under equilibrium conditions.

An increase in maternal arterial $P_{O_2}$ in the ewe was accompanied by very small or negligible changes in $P_{O_2}$ in the other three vessels, whereas in the mares the effect on foetal levels was much greater with either chloralose or sodium pentobarbitone anaesthesia. In both experiments on the mares shown in Fig. 1, there was an increase in umbilical venous $P_{O_2}$ when maternal arterial levels were raised, and the $O_2$ tension in the umbilical vein exceeded that in the uterine vein. When arterial $P_{O_2}$ in the mare returned to near normal, the uterine and umbilical venous $P_{O_2}$ levels converged and under conditions of slight hypoxia (55–60 mm Hg, arterial $P_{O_2}$) umbilical venous $P_{O_2}$ fell below that in the uterine vein. The changes in the umbilical artery of the foetal foal were less marked than those of the vein during alterations in maternal $P_{O_2}$ but there appeared to be a fairly close relationship between the $P_{O_2}$ in the umbilical artery and that in the uterine vein (Fig. 1).

The differences between the two species were emphasized when the data for foetal and uterine venous $P_{O_2}$ from all the experiments in this series (seven ewes and seven mares) were plotted against maternal arterial $P_{O_2}$ on a log. scale (Fig. 2). The observations on the ewes formed a uniform series which showed a rise in foetal and uterine venous $P_{O_2}$ up to 80–100 mm Hg maternal arterial $P_{O_2}$ with little change thereafter. Thus uterine venous levels did not exceed about 60 mm Hg, while values for the umbilical vein remained below 45 mm Hg over the range of 100–400 mm Hg in the maternal arterial blood. A completely different picture was obtained in the mare (Fig. 2). Over the whole range of levels tested log. $P_{O_2}$ in the maternal artery was linearly related to the $P_{O_2}$ in the umbilical artery, the umbilical vein and the uterine vein. However, the rate of rise in umbilical venous $P_{O_2}$ ($b = 73.7 \pm 6.0$) between 28 and 300 mm Hg maternal arterial $P_{O_2}$ was significantly greater ($P < 0.01$) than the corresponding slope for
either uterine venous $P_{O_2}$ ($b = 44.2 \pm 6.3$) or foetal arterial $P_{O_2}$ ($b = 30.0 \pm 4.2$). Furthermore, there was a fairly close parallel between the slope for uterine venous blood and that for foetal arterial blood; the difference between the two was not statistically significant.

Umbilical venous $P_{O_2}$ was equal to that in the uterine vein at a maternal arterial level of about 100 mm Hg; at this point of intersection of the two regression lines, umbilical and uterine venous $P_{O_2}$ was 57 mm Hg. These

absolute figures must, however, be interpreted with some caution since data from mares anaesthetized with sodium pentobarbitone were included in the analysis. $P_{O_2}$ values in these experiments were always higher than with chloralose anaesthesia due to the raised maternal $P_{CO_2}$ levels. It is probable, therefore, that the equilibrium point between umbilical and uterine venous blood would be attained at a maternal arterial $P_{O_2}$ somewhat above 100 mm Hg at normal $P_{CO_2}$ levels.

**The effect of changes in maternal arterial $P_{O_2}$ on uterine venous $P_{O_2}$ in early gestation in the ewe**

The relatively small changes in $P_{O_2}$ in both uterine and umbilical venous blood during oxygen administration to the ewe reported in both past
(Parker & Purves, 1967) and present experiments, were found during the latter part of gestation. However, between 100 days and term the placental circulation is fully developed and uterine blood flow, expressed in terms of unit weight of the uterus and its contents, does not change markedly, whereas in early gestation the cotyledons are small or even rudimentary (D. H. Steven, unpublished observations) and the rate of uterine blood flow per kg uterus is higher than at any other time in gestation (Huckabee, Metcalfe, Prystowsky & Barron, 1961). During the course of the present investigation the effects of changes in maternal arterial $P_{O_2}$ on uterine venous $P_{O_2}$ were also examined in five sheep at 34–60 days gestation. At this stage the foetal vessels were too small to implant catheters without impeding blood flow in the umbilical circulation.

Between 34 and 41 days uterine venous $P_{O_2}$ increased linearly with maternal arterial $P_{O_2}$ which was varied between 50 and 400 mm Hg. This relation is shown in Fig. 3 in which the results from three ewes are plotted as filled circles on a log./log. scale. By 50–60 days the relation between arterial and uterine venous $P_{O_2}$ (open circles) was essentially the same as that found in the latter part of pregnancy (see Fig. 2), in that uterine venous $P_{O_2}$ remained virtually unchanged above arterial levels of 100 mm Hg.

Fig. 3. The relation between the $P_{O_2}$ in maternal arterial blood and that in the uterine vein in five ewes. ●, 34–41 days gestation (three ewes); ○, 50–60 days gestation (two ewes).
The effect of changes in maternal arterial $P_{\text{CO}_2}$ on uterine and umbilical blood gas tensions

Effect on $P_{\text{CO}_2}$ in umbilical and uterine venous blood. The $P_{\text{CO}_2}$ difference between uterine and umbilical venous blood in the ewe under resting conditions was 3–4 mm Hg (Table 1). These observations were made at relatively stable maternal arterial $P_{\text{CO}_2}$ levels (mean $36.3 \pm 0.6$ mm Hg, range 30–40 mm Hg), and the effects of larger changes in $P_{\text{CO}_2}$ in the ewe were investigated in three additional experiments. Previous experience

Fig. 4. The effects of changes in maternal arterial $P_{\text{CO}_2}$ in two ewes anaesthetized initially with halothane, [ ] and later with sodium pentobarbitone, [ ].

(a) $P_{\text{CO}_2}$ changes in maternal artery ( ), uterine vein ( ) and umbilical vein ( ).

(b) pH changes in maternal artery ( ) and umbilical vein ( ). †, hyperventilation with air begun; †, hyperventilation with 5–10% CO$_2$ in air begun; both were continued until the next set of samples had been taken. Further details are given in the text.
had shown that closed-circuit halothane anaesthesia in the ewe invariably resulted in much higher resting blood $P_{CO_2}$ levels than anaesthesia with sodium pentobarbitone (Comline & Silver, 1970). In the present experiments halothane was administered in an $O_2/N_2$ mixture to keep the maternal arterial $P_{O_2}$ between 80 and 110 mm Hg. The $P_{CO_2}$ remained between 55 and 88 mm Hg for long periods without any of the severe respiratory or circulatory changes associated with such high levels of $P_{CO_2}$ under sodium pentobarbitone anaesthesia. Maternal arterial $P_{CO_2}$ was reduced either by hyperventilation with the $O_2/N_2$ mixture or by changing the anaesthetic from halothane to sodium pentobarbitone. The results obtained in two ewes are shown in Fig. 4. In both experiments the change in anaesthetic had a very pronounced effect on resting maternal arterial $P_{CO_2}$ but the $P_{CO_2}$ gradient between uterine and umbilical venous blood did not vary to any extent.

The data for all three experiments have been plotted in Fig. 5 together with observations made at normal maternal $P_{CO_2}$ levels in the seven experiments carried out entirely under sodium pentobarbitone anaesthesia. Both uterine and umbilical venous $P_{CO_2}$ rose linearly with the maternal arterial levels and there was no apparent change in the gradient over the whole range. The mean difference of $4.22 \pm 0.39$ mm Hg between the $P_{CO_2}$ in the two vessels was highly significant ($P < 0.001, n = 47$).

In the mare examination of the data for all experiments showed that,
irrespective of maternal $P_{O_2}$, uterine and umbilical venous $P_{CO_2}$ also rose linearly with an increase in maternal arterial $P_{CO_2}$ (Fig. 5) and there was no apparent change in the gradient between the two vessels over the whole range of maternal levels. In fact, the two series of data were virtually superimposed although analysis of the thirty-six pairs of observations indicated that there was a significant difference of $1.29 \pm 0.46$ mm Hg between them ($P < 0.01$).

Fig. 6. The relation between maternal arterial $P_{CO_2}$ and the $P_{O_2}$ in the uterine vein (○) and umbilical vein (●) in six ewes and seven mares in which maternal arterial $P_{O_2}$ was maintained between 80 and 110 mm Hg at different $P_{CO_2}$ levels.

Effect on $P_{O_2}$ in umbilical and uterine venous blood. Motoyama, Rivard, Acheson & Cook (1967) reported large changes in foetal blood $P_{O_2}$ when maternal acid–base balance was altered in the ewe, but in their experiments maternal $P_{O_2}$ was maintained above 200 mm Hg and the uterine venous $P_{O_2}$ was not measured. In the present investigations $P_{O_2}$ levels in the uterine vein as well as the umbilical vessels were determined during changes in maternal $P_{CO_2}$. All values obtained at normal maternal arterial $P_{O_2}$ levels have been plotted against maternal arterial $P_{CO_2}$ in Fig. 6. As expected, umbilical venous $P_{O_2}$ increased with maternal arterial $P_{CO_2}$ (Motoyama et al. 1967); the changes in $P_{O_2}$ in the uterine vein were similar in magnitude to those in the umbilical vein so that the gradient between the two was maintained at about the same level over the range 15–88 mm Hg $P_{CO_2}$ in the maternal artery.

In the mare both umbilical and uterine venous $P_{O_2}$ levels were affected by changes in maternal arterial $P_{CO_2}$ in a manner similar to that observed in the sheep (Fig. 6).
Table 3. Effect of maternal alkalosis on foetal umbilical pH and $P_{CO_2}$ in five ewes

Mean levels ± s.e.

<table>
<thead>
<tr>
<th>Experimental procedure</th>
<th>No. of observations</th>
<th>Maternal arterial blood</th>
<th>Foetal umbilical blood</th>
<th>Venous</th>
<th>Arterial</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>pH</td>
<td>$P_{CO_2}$ (mm Hg)</td>
<td>pH</td>
<td>$P_{CO_2}$ (mm Hg)</td>
</tr>
<tr>
<td>I Control levels (normal respiration)</td>
<td>*6</td>
<td>7.48 ± 0.010</td>
<td>35.1 ± 1.3</td>
<td>7.37 ± 0.009</td>
<td>43.2 ± 1.4</td>
</tr>
<tr>
<td>II Metabolic alkalosis; i.v. infusion of 5% Na$_2$CO$_3$ into maternal circulation, ½–2½ hr</td>
<td>†9</td>
<td>7.74 ± 0.011</td>
<td>39.5 ± 1.6</td>
<td>7.39 ± 0.010</td>
<td>49.8 ± 1.7</td>
</tr>
<tr>
<td>III Respiratory alkalosis; maternal hyperventilation, ½–2½ hr</td>
<td>‡9</td>
<td>7.71 ± 0.005</td>
<td>18.8 ± 0.5</td>
<td>7.51 ± 0.009</td>
<td>25.6 ± 0.9</td>
</tr>
</tbody>
</table>

* Initial values; one sample from each of four ewes and two samples (separated by ½ hr) from one ewe.
† Three ewes: 2–4 samples from each taken at ½–1 hr intervals.
‡ Two ewes: 4–5 samples from each taken at ½ hr intervals.
**Effect of changes in maternal arterial pH on uterine and umbilical venous pH and gas tensions**

Alterations in maternal arterial $P_{\text{CO}_2}$ were invariably associated with concomitant changes in maternal and foetal pH. Thus the changes in $P_{\text{CO}_2}$ and $P_{O_2}$ in uterine and umbilical venous blood shown in Fig. 5 and 6 could also be closely related to changes in maternal arterial pH (see Fig. 4). Although instances in which foetal pH levels do not follow those of the mother have been reported, there appear to be certain discrepancies. Motoyama *et al.* (1967) found in acute experiments with the foetus exteriorized that metabolic alkalaeemia or prolonged hyperventilation (2–3 hr) in the ewe was associated with a fall in foetal blood pH, whereas Blechner, Meschia & Barron (1960), using conscious sheep with indwelling catheters, suggest that the foetus may be unaffected by large changes in maternal pH. This problem was therefore investigated in a further series of animals under the conditions of our experiments.

In three ewes 5 % $\text{Na}_2\text{CO}_3$ was infused continuously into the maternal circulation at a rate sufficient to maintain the maternal pH above 7.7. This resulted in little or no change in foetal pH whether the infusion was continued for 30 min or as long as 2–3 hr (Table 3). There was, however, a slight compensatory rise in $P_{\text{CO}_2}$ in both maternal and foetal blood under these circumstances. Two other animals were ventilated artificially at a stroke volume sufficient to maintain the maternal arterial $P_{\text{CO}_2}$ below 20 mm Hg for 2½–3 hr. The foetal pH remained high throughout the resultant maternal alkalaeemia (Table 3) and even at the end of the 3 hr period there was no evidence of the metabolic acidosis or other signs of foetal distress described by Motoyama *et al.* (1967) after prolonged maternal hyperventilation.

**DISCUSSION**

$P_{O_2}$ gradients between mother and foetus. The exchange of oxygen across the six layers of the diffuse epitheliochorial placenta of the mare paradoxically appears to be one of the most efficient so far investigated and the tissues of the foetal foal are supplied with blood at a $P_{O_2}$ which is higher than that of any other species for which reliable measurements at normal maternal arterial $P_{O_2}$ are available. In fact, the foetal arterial $P_{O_2}$ of the foal is as high, if not higher, than the umbilical venous $P_{O_2}$ of the lamb, goat, monkey and human, measured under a diversity of anaesthetic and experimental conditions (Metcalf *et al.* 1967).

With the information at present available, the precise reasons for the apparent efficiency of the equine placenta cannot be identified specifically or separated from the many factors known to influence gaseous exchange
in this organ (Metcalf et al. 1967). It seems unlikely that the high foetal
$P_{O_2}$ levels recorded in the foal were due to impairment of umbilical blood
flow, or to any effects of anaesthesia since similar results were obtained
with different anaesthetics and in conscious animals. Furthermore, in the
sheep at least, the presence of catheters within the uterine and umbilical
vessels does not appear to lead to any major changes in umbilical blood
flow in either the conscious (Crenshaw, Huckabee, Curet, Mann & Barron,
1968) or anaesthetized animal (Heymann & Rudolph, 1967).

In the equine placenta the exchange between maternal and foetal blood
occurs in the many thousands of microcotyledons which line the utero-
placental interface. The rapid changes in umbilical $P_{O_2}$ when maternal
arterial $P_{O_2}$ is altered are consistent with a counter-current vascular
arrangement in the microcotyledons and histological evidence supports
this hypothesis, for the arterial blood supply to each cotyledon is arranged
in such a way that the blood in the maternal and foetal capillary networks
appears to flow in opposite directions (Tsutsumi, 1962; Steven, 1968). The
much steeper rise in $P_{O_2}$ of the umbilical vein compared with that of the
uterine vein, which leads to the complete reversal of the $P_{O_2}$ gradient
between the two vessels at high maternal arterial $P_{O_2}$ levels, might be
expected with such an arrangement. In these terms the substantial $P_{O_2}$
gradient between maternal arterial and umbilical venous blood could be
ascribed to the comparatively long diffusion pathway through the inter-
vening tissues and their probable oxygen consumption. On the other hand,
many other factors, such as the perfusion rate in relation to the length of
the capillary bed and the possibility of local or regional alterations in
placental blood flow must also be taken into account.

The close parallel between uterine venous and umbilical arterial $P_{O_2}$
is also consistent with the counter-current hypothesis. However, it can-
not be assumed that mixed uterine venous blood is necessarily represen-
tative of that leaving the exchange areas of the placenta, for the uterus of
the mare contains a thick glandular endometrium and its contribution to
the total effluent blood flow has yet to be measured. Even in the sheep, in
which the intercotyledonal endometrium is poorly developed, the pro-
portion of uterine blood which supplies the cotyledons may vary. Near
term 80–90 % of the total uterine blood flow is distributed to the placenta
(Power, Longo, Wagner, Kuhl & Forster, 1967; Makowski, Meschia,
Droegemueller & Battaglia, 1968), but at 80 days gestation the endometrial
component appears to be much greater, only 63 % of the total flow being
of placental origin (Makowski et al. 1968). Similar figures for placental blood
flow (63–67 %) have been reported for the rabbit near term (Duncan, 1969).
The non-placental component of the uterine circulation may thus vary,
not only with the species but also with gestational age and activity of the
tissue. The flow may also vary between the different areas of the placenta, for Power et al. (1967) found that labelled macroaggregates of albumen were unevenly distributed to the cotyledons of the ewe at normal maternal arterial $P_{O_2}$. They calculated that the more uniform distribution which occurred during hypoxia might lower the $P_{O_2}$ gradient between the uterine and umbilical vein by about 5 mm Hg. This difference is identical with that derived from our data on the sheep (Comline & Silver, 1968) for at high maternal arterial $P_{O_2}$ the gradient between uterine and umbilical venous blood is 18 mm Hg whereas at hypoxic levels the $P_{O_2}$ difference is 13 mm Hg. The mechanisms responsible for the alteration in blood flow to individual cotyledons have not yet been identified, but arterio-venous anastomoses in the maternal blood supply have been reported (Steven, 1966; Makowski, 1968).

Various explanations have been suggested to account for the large transplacental $P_{O_2}$ gradient in the sheep, and the relative constancy of umbilical $P_{O_2}$, particularly during oxygen administration to the mother. It is difficult to reconcile some of these with the findings in the mare, for the relative impermeability of the tissues which separate the foetal and maternal blood (Barron, 1951; Metcalfe, Moll, Bartels, Hilpert & Parer, 1965) and the high degree of oxygen consumption by the placenta itself (Campbell et al. 1966) could equally well apply to the equine placenta. Steven (1968) has put forward a hypothesis based on the very different vascular patterns in the maternal exchange areas, which goes some way to explain these anomalies. In the mare the microcotyledons have a simple capillary bed supplied directly with arterial blood. In contrast a considerable part of the maternal capillary network of each cotyledon of the ewe appears to receive a mixture of arterial blood and blood which has already undergone some exchange. The relative proportions from each source presumably vary in different parts of the cotyledon, but the shape of the $O_2$ dissociation curve will mean that the majority of foetal villi are exposed to a low maternal $P_{O_2}$. In short, while the evidence in the mare points, with some reservations, to a counter-current system, that in the ewe can be explained more easily in terms of the pool system of Metcalfe et al. (1967).

It is tempting to suggest that in the very early stages of gestation in the ewe the transfer of oxygen is more akin to that in the mare, since the cotyledons are relatively undeveloped, the maternal capillary networks are simple (D. H. Steven, unpublished observations) and the $P_{O_2}$ of the uterine venous blood is extremely sensitive to changes in arterial levels. The similarity may, however, be more apparent than real, since preliminary observations on the $P_{O_2}$ in umbilical blood at 40 days gestation in the ewe indicate that even at this early stage the foetal $P_{O_2}$ does not rise above that
found later in gestation when maternal levels are increased. Furthermore, the considerable development of the endometrial vasculature in early pregnancy suggests that the blood flow to these regions must be high, perhaps exceeding that in the cotyledons; indeed the foetus may take second place to developing tissues of the uterus at this time. The present findings on the relation between arterial and uterine venous $P_{O_2}$ in early pregnancy would certainly indicate a marked change in conditions between 40 and 50 days, at the time when uterine blood flow per unit weight falls most steeply (Huckabee et al. 1961). These changes may well be associated with alterations in the hormonal climate; in fact very similar differences in uterine blood flow have been demonstrated for the non-pregnant uterus at different stages of the oestrous cycle of the ewe (Huckabee, Crenshaw, Curet & Barron, 1968).

$P_{CO_2}$ and pH in maternal and foetal blood. Since $CO_2$ is more rapidly diffusible than $O_2$ the presence of much smaller $P_{CO_2}$ gradients from foetus to mother in both ewe and mare was not unexpected. Indeed, on a number of occasions $P_{CO_2}$ levels in the umbilical vein of the foetal foal were slightly lower than those in the uterine vein. This situation was never encountered in the ewe, which again emphasizes the underlying differences in blood gas transfer between the two species.

The changes in resting $P_{O_2}$ levels which were observed in the foetal blood and in the uterine vein during respiratory alkalaeemia or acidaeemia, in both mare and ewe, could most readily be attributed to the effects of $CO_2$ and pH changes on the $O_2$ dissociation curves for haemoglobin. In a number of species, including the ewe, a pronounced Bohr effect may be produced by a change of 0-1–0-2 pH units in both foetal and maternal blood (Bartels & Metcalfe, 1965). However, other factors such as uterine and umbilical blood flow may be affected by alterations in maternal acid–base balance. In the present experiments alterations in maternal pH of respiratory origin whether of short or long duration were always followed by similar changes in pH in the foetus, and thus $P_{O_2}$ levels in foetal and uterine venous blood were all shifted in the same direction. In view of these findings, it is difficult to interpret the curious effects of prolonged hyperventilation (with hypocapnia) found by Motoyama et al. (1967). It may be that a drastic fall in umbilical blood flow and metabolic acidosis of the foetus is more liable to occur with this procedure when the foetus is removed from the uterus.

The suggestion, put forward by Blechner et al. (1960) that changes in foetal and maternal pH of metabolic origin may, on occasions, be completely independent of one another, is amply borne out in the present experiments on ewes with infusions of $Na_2CO_3$. It would appear that neither this nor NH$_4$Cl (Blechner et al. 1960) are freely diffusible from
mother to foetus. Experimental alteration of foetal pH was not attempted, but it is well known that after anoxia the low pH, due to increased lactate production in the foetus, may persist for many hours. Alexander, Britton, Cohen & Nixon (1969) have suggested that the placenta of the ewe is virtually impermeable to lactate. No information on the passage of fixed acids and bases across the equine placenta is available but the problem merits further investigation in view of the apparently greater efficiency of blood gas transfer in this species.

Effects of removal of the foetus from the uterus. A comparison of the present findings on conscious and anaesthetized animals shows that similar results can be obtained under very different conditions. A common factor in all the experiments was the relatively slight disturbance of the foetus which remained within the uterus. This suggests that the variations in the absolute values of several components of the foetal blood, particularly the $P_{O_2}$ which can be found in the literature, may be due to the removal of the foetus, partially or completely, from the uterus. Heymann & Rudolph (1967) found that this procedure caused a significant rise in umbilical venous $P_{O_2}$, which they ascribed primarily to a fall in umbilical blood flow. Changes in the circulation to different regions of the foetus (Campbell, Dawes, Fishman & Hyman, 1967) and a decrease in its oxygen consumption (Meschia, Cotter, Makowski & Barron, 1967) no doubt also contribute to the effect although the importance of these factors will depend on the maturity of the foetus and the depth and type of anaesthesia. In addition, the effect on uterine haemodynamics of the reduction in its volume through the loss of amniotic fluid as well as the foetus has, as yet, received little attention. The surprisingly close agreement between the values from anaesthetized ewes and mares and those from conscious animals in the present experiments indicates that anaesthesia per se has little apparent effect on foetal blood gas levels, provided care is taken to keep maternal $P_{O_2}$, $P_{CO_2}$, pH and blood pressure within normal limits.

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REFERENCES


