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Ammonia Production by the Pregnant Uterus[1] (39868)

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Introduction. The major emphasis of investigations of nitrogen metabolism of the uterus and conceptus has been on the anabolic requirements for the synthesis of new tissues (1, 2). In late gestation, however, the lamb fetus excretes relatively large quantities of urea (3). This observation has prompted us to examine further the catabolic processes of gestation and to seek evidence concerning the role of ammonia as an excretory product of the pregnant uterus.

Materials and Methods. Two nonpregnant and seventeen pregnant Western-breed ewes with gestational ages of 40 to 130 days were fasted for 48 hr and then placed under pentobarbital sedation (5 mg kg^{-1}) and spinal anesthesia (6 mg of pontocaine in hyperbaric glucose). Polyvinyl catheters were placed in the maternal femoral artery and both uterine veins (4). Umbilical venous and pedal arterial catheters were implanted in seven fetuses. Three other fetuses were studied at the time of Cesarean section by catheterization of the umbilical vein and artery. All lambs were appropriately grown at the time of delivery. Arterial blood samples were obtained from two nonpregnant sheep.

Simultaneous fetal and maternal blood samples were obtained each morning after a recovery period of at least 48 hr. On the average, each animal was studied 10 times over a period of 14 days. Blood samples for ammonia determinations were withdrawn into dry, 3-ml plastic syringes containing EDTA; those for measurement of oxygen content were drawn into heparinized capillary tubes. Whole blood (0.5 ml) was analyzed immediately for ammonia, using the method of Kurahasi *et al.* (5). Oxygen contents were determined with a Lex-O_2-Con (Lexington Instrument Corp.) calibrated with distilled water saturated with oxygen at 0°.

All ammonia samples were analyzed in triplicate with a coefficient of variation of 3.9%. Paired *t* tests showed no significant differences between venous samples from the left and right uterine horns of each ewe in either ammonia concentrations or oxygen contents. Therefore, a mean venous concentration was used in subsequent calculations.

Results. The between-group differences in mean arterial ammonia concentrations (fetus, 49.27 ± 2.35 μM; pregnant ewe, 36.70 ± 0.93 μM; nonpregnant ewe, 23.04 ± 1.01 μM) were statistically significant ($P < 0.001$) (Fig. 1). All the veno-arterial differences in ammonia concentration across the uterine circulation were positive (Fig. 2a), i.e., there was a consistent excretion of ammonia from the gravid uterus into the maternal circulation. The veno-arterial difference was considerably higher (64.85 ± 3.27 μM) at <80 days of gestation than at >80 days of gestation (25.85 ± 0.77 μM, SEM). The simultaneous measurement of ammonia and oxygen veno-arterial differences permits a comparison of ammonia production rate to the rate of oxygen consumption (Fig. 2b). Clearly, during early gestation there is a higher ammonia production rate by the gravid uterus in relation to oxygen consumption than during late gestation.

The veno-arterial differences of ammonia concentration across the umbilical circulation of the fetus from 114 days until term were all positive, with a mean ± 1 SE of 13 ± 1.4 μM. This difference is significantly different from 0 by the sign test ($P < 0.01$).

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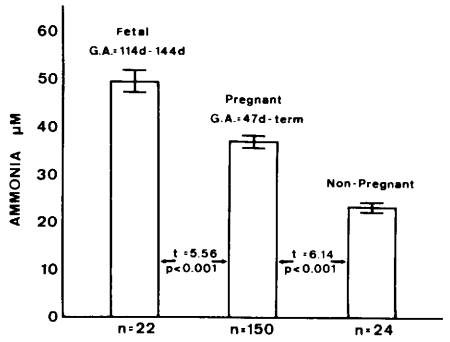

FIG. 1. Arterial ammonia concentrations in fetal, pregnant, and nonpregnant sheep. Concentrations of ammonia are given as micromoles (mean ± SE).

Thus, ammonia is delivered into *both* the uterine and umbilical circulations from tissues of the uterus other than the fetus.

Discussion. It is apparent that the pregnant ovine uterus excretes ammonia into the maternal circulation throughout pregnancy. The source of this ammonia appears to be the placenta, because the umbilical circulation also shows a net uptake of this substance. It should be noted that at 2 months of gestation the ovine placenta is the largest part of the conceptus and has achieved its maximal size, weighing ~400 g, whereas the fetus weighs ~60 g. Hence, the finding that ammonia production is maximal, when compared with oxygen uptake, between 60 and 70 days of gestation could be related to the fact that placental metabolism is a large fraction of total uterine metabolism in early pregnancy. *In vitro* studies of slices of human placenta have shown it is capable of producing ammonia (6, 7).

Quantitatively, the uterine excretion of ammonia in early pregnancy is an important phenomenon. At 70 days the pregnant uterus weighs ~1.16 kg (fetal fluid excluded), contains 14.17 g of nitrogen kg^{-1} of tissue, and grows at the rate of ~40 g day^{-1} (1). Thus, nitrogen accumulation in the form of new tissue is ~0.57 g day^{-1}. Oxygen consumption of the gravid uterus at 70 days equals ~0.42 mmole min^{-1} (8). Given this rate of oxygen uptake and the ammonia/oxygen ratios presented in Fig. 2b, it can be estimated that the nitrogen excreted by the uterus, in the form of ammonia, is ~0.45 g day^{-1}. Therefore, at 70 days of gestation the ratio of nitrogen excreted by the uterus as ammonia to the combined total nitrogen requirements for both growth and ammonia excretion is ~44%.

The excretion of ammonia from uterus to maternal blood can also be estimated for the last 20 days of gestation (125 to 145 days), when the uterine venoarterial difference of ammonia is fairly constant. Mean uterine oxygen uptake is 0.36 mmole min^{-1} kg^{-1} (8), and the mean uterine ammonia/oxygen ratio is 15.8 μmol $mmole^{-1}$ (Fig. 2b). Therefore, late in pregnancy, a gravid uterus that weighs 4.5 kg excretes ~25.6 μmole min^{-1} or 0.5 g day^{-1} of ammonia nitrogen. At this stage of gestation, the fetus weighs ~3.5 kg and has nitrogen requirements for growth and urea production equal to ~3.5 g day^{-1} (2). Thus, toward the end of pregnancy, ammonia production by the uterus represents a smaller fraction (~12%) of nitrogen requirements.

The fetus receives ammonia from the placenta. The fetal uptake of ammonia, calculated as the product of mean umbilical veno-arterial difference (0.013 μmole ml^{-1}) times mean umbilical blood flow (175 ml min^{-1} kg^{-1}) (9), is ~2.3 μmol min^{-1} kg^{-1} or 0.046 g day^{-1} kg^{-1} of nitrogen. This is an insignificant amount in comparison with the quantity of nitrogen received by the fetus in the form of amino acids (2), but it is probably an important factor in determining the relatively high level of ammonia in fetal blood. Similarly, the production rate of ammonia by the pregnant uterus could readily explain the 12-μmole mean difference of ammonia concentration between pregnant and nonpregnant ewes, because it would require an ammonia clearance of 2 liters min^{-1} in order to clear 24 μmol of ammonia per minute with a concentration increment of 12 μmole.

The data presented demonstrate for the first time that ammonia is an important metabolic end product of the ovine gravid uterus, especially in early pregnancy, and that the placenta is the probable site of ammonia formation. It would seem that the ovine conceptus (i.e., placenta and fetus) has a developmental pattern of nitrogen excretion similar to that described in amphibians and birds (10), namely, an initial am-

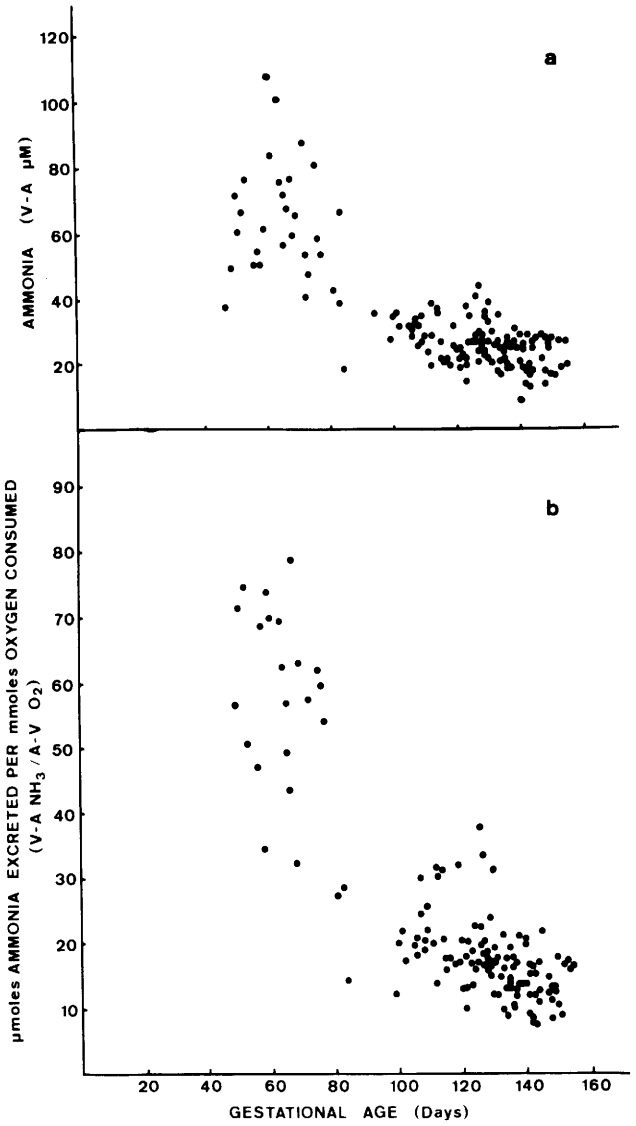

FIG. 2. (a) Difference in ammonia concentration (micromoles) between uterine venous and femoral arterial blood in pregnant sheep from 47 days to term. (b) Ammonia (in micromoles) excreted into the maternal circulation per millimole of oxygen consumed by the gravid sheep uterus from 47 days to term.

moniotelic phase followed by a period in which the ureotelic or uricotelic mode becomes predominant.

Summary. The arterial-venous differences of ammonia and oxygen were measured across the uterine and umbilical circulations in nonanesthetized, chronic sheep preparations from 47 to 155 days of gestation. In all cases, ammonia was excreted from the uteroplacental unit into both the uterine and umbilical circulations. In early gestation (70 days), the gravid uterus excretes into the maternal blood ~55 μmol of ammonia/mmole of oxygen consumed and it can be estimated that the ratio of nitrogen excreted by the uterus as ammonia to the combined total nitrogen requirements for both growth and ammonia excretion is ~44%. Late in gestation, the gravid uterus excretes ~16 μmol of ammonia/mmole of oxygen consumed. This represents ~12% of fetal nitrogen requirements.

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