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Title Cardiovascular Changes During Pregnancy in the Ewe

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In order to study maternal cardiovascular adjustments to pregnancy, serial determinations were carried out on five pregnant and four nonpregnant ewes. Measurements were made at approximately two to three week intervals, and continued for nine weeks after delivery.

Cardiac output was measured with the dye dilution technique using Evans blue dye and a continuously recording densitometer. Plasma volume was simultaneously estimated. Intra-arterial blood pressure was measured with a strain gauge transducer. All measurements were made on the sheep while they stood quietly in a stanchion, only local anesthesia being used at the sites of vessel puncture. Carotid loops were constructed to facilitate arterial catheterization.

Statistical analysis was carried out by subtracting the average value of each function in each sheep for 30 days after delivery

from the average value of the same function for 30 days before delivery. The differences obtained in the pregnant sheep were compared with figures similarly obtained in the nonpregnant sheep by means of the t-test.

The average values of the measurements in the pregnant ewes in the last three weeks of pregnancy were as follows:

a. Cardiac output, 10.3 lit/min. This was 41% greater than the average postpartum value, and the difference was statistically significant ($P < 0.01$).

b. Cardiac output per unit of body weight, 157 ml/kg/min. This was 31% greater than the postpartum average ($P < 0.02$).

c. Heart rate, 109 beats per minute. This was 30% greater than the postpartum average ($P < 0.02$).

d. Arterial systolic blood pressure, 88 mm Hg. This was 18% less than the postpartum average ($P < 0.1$).

e. Arterial diastolic blood pressure, 74 mm Hg. This was 15% less than the postpartum average (N.S.).

f. Peripheral resistance, 616 dyne sec cm^{-5}. This was 42% less than the postpartum average ($P < 0.01$).

g. Blood volume, 4.9 liters. This was 9% greater than the postpartum average ($P < 0.05$).

h. Blood volume per unit of body weight, 74 ml/kg. This was 1% greater than the postpartum average (N. S.).

i. Plasma volume, 3.4 liters. This was 10% greater than the postpartum average ($P < 0.05$).

j. Hematocrit, 31%. This was the same as the postpartum average (N. S.).

k. Body weight, 66.8 kg. This was 8% greater than the postpartum average ($P < 0.01$).

The changes accompanying pregnancy in the ewes were similar in many respects to those reported for the human. The main differences noted were the smaller increase in blood volume and absence of "anemia of pregnancy" in the sheep. The possible distribution of the increased blood flow and mechanisms involved in the circulatory adjustments to pregnancy are discussed.

CARDIOVASCULAR CHANGES DURING PREGNANCY IN THE EWE

by

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TABLE OF CONTENTS

TABLE OF CONTENTS

LIST OF FIGURES

LIST OF TABLES

CARDIOVASCULAR CHANGES DURING PREGNANCY IN THE EWE

I INTRODUCTION

During the course of pregnancy, the mammalian fetus is entirely dependent on its mother for nutrient supply and waste removal. More specifically, it is dependent on the maternal blood circulatory system. In order to supply these needs, profound changes occur in the maternal circulation as gestation proceeds. It seems likely that the extent and type of these changes will have significance for both mother and fetus. For instance, an increase in the work of the heart could be deleterious to unhealthy mothers. On the other hand, in some cases the circulatory adjustments may be inadequate to meet the needs of the growing fetus, and fetal development or health may be impaired. In order to define the conditions necessary for normal fetal growth, there is a need to know what the maternal circulatory adjustments to pregnancy are, and also the mechanisms by which they arise.

This report consists of a review of the published literature on this subject, and also the results of an experiment evaluating the circulatory changes occurring in ewes during pregnancy.

II REVIEW OF THE LITERATURE

Most of the experimental work concerning the circulation during pregnancy in the clinically healthy mother has been carried out in the human. This review presents the major findings of these reports, and for convenience tabulations are used. When using tables certain simplifications must be made in order to accommodate data from experiments of different design, and this in its turn imposes restrictions on the worth of the table. These restrictions will be brought out in the discussion of the tables below.

Some of the work goes back almost half a century and it is recognized that many techniques and experimental designs have improved since then. Probably the selection of healthy subjects has also become more precise. However, despite these limitations, the early work opened up the way with important general findings, and laid the foundation for the more refined and detailed work which followed.

Cardiac Output

The cardiac output is the quantity of blood ejected by one ventricle of the heart per minute. Table 1 contains a summary of reports of the changes in cardiac output occurring during pregnancy in humans and dogs. Control values representing the cardiac output

TABLE 1 CARDIAC OUTPUT DURING PREGNANCY

Control value (L/min)	Extreme value in preg. (L/min)	Max. change (%)	Time of occurrence of extreme (weeks)	Method	Reference
HUMANS					
3.4 prepreg.	5.2	53	-	N_2O	Lindhard 1915 (45, p. 314-316)
3.8 nonpreg.	6.2	63	-	N_2O	Weiss 1924 (76)
4.4 postpart.	5.7	30	-	N_2O	Gammeltoft 1926 (25)
3.8 nonpreg.	5.3	40	-	N_2O	Haupt 1927 (34)
3.8 postpart.	8.0	110	-	Acetylene	Stander and Cadden 1932 (66)
3.2 postpart.	5.2	62	30	Acetylene	Burwell et al. 1938 (13, p. 986-988)
5.0 postpart.	5.8	16	-	Fick O_2	Palmer and Walker 1949 (56)
4.5 nonpreg.	5.7	27	-	Fick O_2	Hamilton 1949 (28)
2.4 nonpreg.	4.8	100	-	Dye dilution	Bucht 1951 (10, p. 45)
6.5 nonpreg.	7.8	20	-	Fick O_2	Werkö 1954 (77, p. 172)
6.3 postpart.	8.3	32	28	Dye dilution	Adams 1954 (2, p. 751)
3.8 postpart.	6.2	63	24-28	Arterial blood pressure analysis	Brehm and Kindling 1955 (9, p. 700-701)
5.0 nonpreg.	7.0	40	-	Fick O_2	Rose et al. 1956 (61, p. 237)
Average increase:		50			
DOGS					
2.9 prepreg.	4.1	41	-	Fick O_2	Stander et al. 1926 (67)

of subjects in the nonpregnant state are given for each experiment. "Prepreg." indicates that the control values were recorded in the subjects before pregnancy began. "Postpart." means that the control values were recorded on the subjects after delivery, in all cases at least six days after, and generally much later than this. "Nonpreg." control values are those recorded on normal nonpregnant subjects by the same techniques, but different subjects were used for the measurements during pregnancy. Clearly there can be objections regarding the validity of these latter control values, particularly if the number of subjects is small. These objections concern the quantitative interpretation of the results rather than their qualitative nature.

The mean highest value of cardiac output obtained during pregnancy is shown for each experiment (Table 1). Where serial measurements have been made on a number of subjects throughout pregnancy, the extreme values are probably close to the true maxima. However, if different subjects have been studied only once in pregnancy and the cardiac output plotted against time during gestation, the curve is unlikely to give a true picture unless numbers are great because of the subject to subject variation in this function. The use of large numbers has generally not been possible in human experimentation because of the complicated nature of the

measurements. Thus experiments where single determinations were made on each subject are likely to show great variability. Also any details of the curve of cardiac output during pregnancy are likely to be missed with small numbers of subjects.

A further restriction on accuracy is imposed by the techniques used to measure cardiac output. Some of the earlier methods gave consistently lower results than the later ones, e.g. the acetylene compared with the direct Fick method (Hamilton, 30, p. 572). However, this should not affect the percent changes in cardiac output (third column of Fig. 1), but only the absolute values.

The mean maximum percent change obtained by averaging the figures in the third column (without weighting for numbers of subjects because of the diversity of experimental design) is 50% for the human. If we consider only those experiments where serial measurements were taken on a number of women during pregnancy (Burwell et al., 14, p. 986-988; Adams, 2, p. 751; Brehm and Kindling, 9, p. 700-701) the mean increase is 52%. In these three studies where serial measurements were made on the same women, it was noted that the maximum value did not occur at term, but some time before, generally at the seventh of the ten lunar months of human pregnancy. After this time the cardiac output declined, tending towards but not reaching control values. This decline in

cardiac output in the last weeks of pregnancy had been noted by other workers. However, it is felt that one could be confident that it was not an artifact only by taking serial measurements on the same women during pregnancy. Burwell et al. (14, p. 982-990) studied this decline of cardiac output before term in women under basal conditions and concluded that it is a real feature of the circulation which is not due to some external factor such as reduced activity on the part of the mother as term approaches. One result of the decline in cardiac output is that the heart work is reduced, and a previously tested reserve is available for the stresses of labor and delivery (27).

Cardiac output during pregnancy was measured in two bitches (Stander, Duncan and Sisson, 67) and an increase of 41% above control values was found. The data are not sufficient to determine if the maximum value occurred some weeks before term, as it does in the human.

One can conclude from this review that in the human during pregnancy the cardiac output increases to a maximum of about 50% above nonpregnant levels. This maximum occurs between the sixth and eighth lunar months, and then declines towards control levels. A similar increase in cardiac output was found during pregnancy in dogs.

Heart Rate and Stroke Volume

The cardiac output consists of the product of heart rate and stroke volume, and it is of interest to note the contribution of each of these components to the cardiac output during pregnancy. The heart rate (Table 2) increased during pregnancy in all the experiments on humans except that of Adams (2, p. 751) who reported no change. The average reported maximum increase was approximately 19%. As with cardiac output, heart rate usually reached a maximum two to three months before term, and declined thereafter. In Brehm and Kindling's (9, p. 699) report however, the heart rate remained fairly constant for the last two months of pregnancy.

Stroke volume (Table 3), calculated from cardiac output and heart rate, increased in all experiments except that reported by Werkö (77, p. 172), where a decrease occurred. Werkö's controls, however, were nonpregnant subjects, and may not have been representative of the subjects used for the measurements during pregnancy. The average maximum change in all experiments was an increase in stroke volume of 15%, and this, like the cardiac output, occurred some months before term, and then declined. The stroke volume, being calculated from the cardiac output, suffers from the same reservations with respect to absolute values. However, the relative change due to pregnancy should not be seriously affected by this.

TABLE 2 HEART RATE DURING PREGNANCY

Control value (per min)	Extreme value in preg. (per min)	Max. change (%)	Time of occurrence of extreme (weeks)	Occurrence of decline from extreme	Reference
HUMANS					
75 postpart.	87	16	29-33	decline	Hare and Karn 1929 (33, p. 387)
70 postpart.	81	16	-	-	Cohen and Thomson 1936 (18, p. 618)
75 postpart.	95	27	28	decline	Landt and Benjamin 1936 (44, p. 597)
75 postpart.	94	25	30	decline	Burwell et al. 1938 (14, p. 984)
68 nonpreg.	82	21	-	-	Hamilton 1949 (28)
-	-	0	-	-	Adams 1954 (2, p. 751)
66 nonpreg.	83	26	-	-	Werkö 1954 (77, p. 172)
70 postpart.	86	23	32-40	no decline	Brehm and Kindling 1955 (9, p. 699)

Average increase:		19			

TABLE 3 STROKE VOLUME DURING PREGNANCY

Control value (ml)	Extreme value in preg. (ml)	Max. change (%)	Time of occurrence of extreme (weeks)	Occurrence of decline from extreme	Reference
HUMANS					
62 postpart.	71	15	-	-	Gammeltoft 1926 (25)
43 postpart.	55	28	30	decline	Burwell et al. 1938 (14, p. 984)
66 nonpreg.	70	6	-	-	Hamilton 1949 (28)
92 postpart.	98	7	28	decline	Adams 1954 (2, p. 751)
100 nonpreg.	84	-16	-	-	Werkö 1954 (77, p. 172)
52 postpart.	79	52	24-28	decline	Brehm and Kindling 1955 (9, p. 699)
Average increase:		15			

Arterial Blood Pressure

There is not complete agreement amongst the various workers regarding the changes occurring in arterial systolic blood pressure during pregnancy (Table 4). Two workers reported no change, and three reported a decrease. The average maximum decrease reported is approximately 6%, and when a decrease occurred, the minimum value was at the eighth or ninth lunar month.

All workers agreed that the arterial diastolic pressure declined during pregnancy, the mean maximum decrease being 13% below control levels. This lowest value occurred variously between the fifth and the ninth lunar month, and then increased as term approached (Table 4).

Adams (2, p. 754) reported that the mean arterial blood pressure decreased to 74 mm Hg at the ninth month of pregnancy, this being 24% lower than the mean postpartum value of 98 mm Hg. The blood pressure then rose to term. Adams carried out serial measurements on a number of women during and after pregnancy.

The fact that diastolic pressure always decreased, and that systolic pressure either did not change or fell, suggests that the mean arterial blood pressure decreased during pregnancy, reaching a minimum value between the fifth and the ninth month, and then increased gradually to term.

TABLE 4 ARTERIAL BLOOD PRESSURE DURING PREGNANCY

Control value (mm Hg)	Extreme value in preg. (mm Hg)	Max. change (%)	Time of occurrence of extreme (weeks)	Occurrence of rise from extreme	Reference
HUMANS		Systolic blood pressure			
-	-	0	-	-	Hare and Karn 1929 (33, p. 387)
112 postpart.	101	-10	36	rise	Landt and Benjamin 1936 (44, p. 596-597)
115 postpart.	-	0	-	-	Burwell et al. 1938 (14, p. 984-987)
121 nonpreg.	106	-12	-	-	Hamilton 1949 (28)
115 postpart.	108	-6	32	rise	Brehm and Kindling 1955 (9, p. 700-701)
Average decrease:		6			
HUMANS		Diastolic blood pressure			
75 postpart.	70	-7	21-25	rise	Hare and Karn 1929 (33, p. 387)
72 postpart.	58	-19	36	rise	Landt and Benjamin 1936 (44, p. 596-597)
80 postpart.	62	-23	30	rise	Burwell et al. 1938 (14, p. 984-987)
75 nonpreg.	70	-7	-	-	Hamilton 1949 (28)
78 postpart.	70	-10	24-28	rise	Brehm and Kindling 1955 (9, p. 700-701)
Average decrease:		13			

Peripheral Resistance

The peripheral resistance in its simplest form is expressed as the mean arterial blood pressure divided by the cardiac output. It is a measure of the pressure developed by the left ventricle when pumping a certain quantity of blood. In all reports where the necessary measurements have been made, the peripheral resistance decreased to the sixth or seventh month, and then increased (Table 5). The decrease in peripheral resistance is considerable, averaging 30%, and it is one of the most striking features of the circulatory changes during pregnancy. Possible mechanisms of this change will be considered in the discussion below.

Blood Volume

In the human (Table 6) the blood volume increased approximately 27% during pregnancy, generally reaching a maximum between the seventh and ninth month. Three of the five authors who carried out serial measurements on the same women during pregnancy noted a decline in blood volume towards control levels after this time, the other two noting no change. In two experiments (Dieckmann and Wegner, 20, p. 83; Roscoe and Donaldson, 60, p. 532), the control values used were measurements made in early pregnancy, and these were probably somewhat above the nonpregnant control values of the same subjects. Thus the value of 27% as the

TABLE 5 PERIPHERAL RESISTANCE DURING PREGNANCY

Control value (dyne sec cm^{-5})	Extreme value in preg. (dyne sec cm^{-5})	Max change (%)	Time of occurrence of extreme (weeks)	Occurrence of rise from extreme	Reference
HUMANS					
-	-	decrease	-	-	Burwell et al. 1938 (14, p. 984-988)
-	-	decrease	-	-	Hamilton 1949 (28)
1080 postpart.	897	-17	28	rise	Adams 1954 (2, p. 754-755)
2100 postpart.	1200	-43	24-28	rise	Brehm and Kindling 1955 (9, p. 700-701)
Average decrease		30			

TABLE 6 BLOOD VOLUME DURING PREGNANCY

Control value (liters)	Extreme value in preg. (liters)	Max. change (%)	Time of occurrence of extreme (weeks)	Occurrence of decline from extreme	Method	Reference
HUMANS						
4.2 postpart.	5.5	31	-	-	Vital red	Miller et al. 1915 (52)
- early preg.	-	23	-	-	Vital red	Dieckmann and Wegner 1934 (20, p. 83)
4.0 postpart.	5.7	42	34	decline	Evans blue	Thomson et al. 1938 (72, p. 51-55)
4.3 early preg.	5.4	26	-	-	Evans blue	Roscoe and Donaldson 1946 (60, p. 532)
4.2 postpart.	5.3	26	-	-	Evans blue	McLennan and Thouin 1948 (49, p. 194-195)
4.5 postpart.	6.1	35	36	decline	Evans blue	Tysoe and Lowenstein 1950 (73, p. 1192)
4.15 nonpreg.	5.14	24	-	-	Evans blue	White 1950 (78)
5.3 nonpreg.	6.3	19	-	-	Evans blue	Bucht 1951 (10, p. 45)
4.2 postpart.	5.85	40	29-34	decline	R.A. Fe	Caton et al. 1951 (16, p. 1214-1215)
4.5 postpart.	5.0	11	34	no decline	Evans blue	Adams 1954 (2, p. 752)
5.0 postpart.	6.2	24	37-40	no decline	-	Werkö 1954 (77, p. 169-170)
Average increase:		27				

TABLE 6 BLOOD VOLUME DURING PREGNANCY (continued)

Control value (liters)	Extreme value in preg. (liters)	Max. change (%)	Time of occurrence of extreme	Occurrence of decline from extreme	Method	Reference
COWS						
-	-	increase	-	-	Vital red	Miller 1932 (53)
-	-	increase	-	-	Evans blue	Reynolds 1953 (59)
SHEEP						
2.4 early preg.	2.9	24	term	no decline	Evans blue	Barcroft et al. 1939 (3)
RATS						
- early preg.	-	41	-	-	Vital red	Bond 1948 (7)
RABBITS						
- prepreg.	-	6	25/32 days	decline	Evans blue	Horger and Zarrow 1957 (37)

average maximum increase in blood volume is probably a little too low.

Although body weight data on the subjects used in these experiments are not generally available, it is improbable that the body weight increase during pregnancy exceeded the control value by more than 27%, i. e. the amount by which the blood volume increased. Much of this increase is due to the weights of the fetus, fetal membranes and fetal fluids. Evans blue, used in most of the blood volume determinations, does not cross the human placenta, so only the maternal blood volume is measured by it and not the blood contained in the fetal mass. Therefore the blood volume per unit of maternal body weight alone (excluding fetal mass) probably increases and a hypervolemia occurs.

The blood volume of cows is reported to increase during pregnancy (Reynolds, 59), and there is also a report by Barcroft, Kennedy and Mason (3) of the blood volume of sheep increasing 24% during pregnancy, reaching a maximum value at term. Barcroft et al. used a control group of nonpregnant ewes to evaluate any seasonal or environmental changes which may have occurred during the period of measurements made on the pregnant sheep. The control values used for their pregnant sheep were made in very early pregnancy, approximately ten days after mating. Regarding other

species, blood volume of rats increased 41% during pregnancy, while that of rabbits increased by only 6% above control values (Table 6).

Plasma Volume

The change in plasma volume parallels that in blood volume during human pregnancy, except that plasma volume increases by a greater amount, reaching an average maximum of 38% above control levels (Table 7). As with blood volume, it reaches its maximum value at the seventh to ninth month, and generally declines to term. In Barcroft et al.'s study with sheep (3), the increase in plasma volume was also greater than the increase in blood volume. That is, most of the increase in blood volume was due to an increase in plasma, rather than cellular volume.

Hytten and Paintin (41) made an important contribution when they determined the variability of the maximum plasma volume increase occurring in a standard group of pregnant women. In the women studied, plasma volume increased 25 to 80%, with an average increase of 46% above control levels. They found that the maximum increase was related to the birth weight of the baby, but not to the size of the mother. This supports the suggestion that the changes occurring in the maternal circulation are brought about through the mediation of the fetus.

TABLE 7 PLASMA VOLUME DURING PREGNANCY

Control value (liters)	Extreme value in preg. (liters)	Max. change (%)	Time of occurrence of extreme (weeks)	Occurrence of decline from extreme	Method	Reference
HUMANS						
2.6 postpart.	3.52	34	-	-	Vital red	Miller et al. 1915 (52)
- early preg.	-	25	-	-	Vital red	Dieckmann and Wegner 1934 (20, p. 83)
2.55 postpart.	3.9	53	34	decline	Evans blue	Thomson et al. 1938 (72, p. 51-55)
2.65 early preg.	3.45	30	-	-	Evans blue	Roscoe and Donaldson 1946 (60, p.532)
2.5 postpart.	3.3	32	-	-	Evans blue	McLennan and Thouin 1948 (49, p. 194-195)
2.8 postpart.	4.2	49	35	decline	Evans blue	Caton et al. 1949 (16, p.477)
2.8 postpart.	4.1	44	36	decline	Evans blue	Tysoe and Lowenstein 1950 (73, p. 1192)
2.36 nonpreg.	3.19	35	-	-	Evans blue	White 1950 (78)
3.4 nonpreg.	4.3	26	-	-	Evans blue	Bucht 1951 (10, p. 45)
2.9 postpart.	4.1	41	29-34	decline	R.A. Fe	Caton et al. 1951 (16, p. 1214-1215)
2.5 postpart.	3.4	36	32-34	no decline	Evans blue	Adams 1954 (2, p. 152-153)
2.7 postpart.	3.9	46	34	decline	Evans blue	Hytten and Paintin 1963 (41)
Average increase:		38				

TABLE 7 PLASMA VOLUME DURING PREGNANCY (continued)

Control value (liters)	Extreme value in preg. (liters)	Max. change (%)	Time of occurrence of extreme	Occurrence of decline from extreme	Method	Reference
COWS						
-	-	increase	-	-	Vital red	Miller 1932 (53)
-	-	increase	-	-	Evans blue	Reynolds 1953 (59)
SHEEP						
1.5 early preg.	2.1	40	term	no decline	Evans blue	Barcroft et al. 1939 (3, p.163-164)
RABBITS						
-	-	12	25/32 days	decline	Evans blue	Horger and Zarrow 1957 (37)

Hematocrit

The hematocrit, i. e. the percent of cellular volume of the blood, decreases during pregnancy in the human, reaching a minimum about the eighth lunar month (Table 8). The red cell mass actually increases during pregnancy by about 17% (Hytten and Duncan, 40, p. 859), but with the concomitant increase in plasma volume of about 40%, there is a decrease in hematocrit. This has been recognized for a number of years and has been called the "anemia of pregnancy". On the average, the minimum value reached is 11% below control levels in the human. This decrease in hematocrit has been recorded in the rat and rabbit, but appears to be absent in the cow (Table 8). Barcroft et al. (3) noted a decrease of 24% in hematocrit in their sheep at term.

The Mechanisms Involved in the Cardiovascular Changes in Pregnancy

The maternal cardiovascular adjustments to pregnancy are rather complex, and a number of theories have been suggested to account for them. Burwell (12; 14, p. 1000) introduced the idea that the pregnant uterus is an area of low vascular resistance which is similar in many ways to an arteriovenous fistula. Some of the similarities between the two conditions that he noted were: increased cardiac output, increased blood volume, increased arterial pulse pressure and elevated venous pressure adjacent to the fistula, i. e. in

TABLE 8 HEMATOCRIT CHANGES ASSOCIATED WITH PREGNANCY

Control value (%)	Extreme value in preg. (%)	Max. change (%)	Time of occurrence of extreme (weeks)	Occurrence of rise from extreme	Reference
HUMANS					
38 postpart.	35.8	-6	-	-	Miller et al. 1915 (52)
35 postpart.	31.5	-10	-	-	Plass and Bogert 1924 (58)
- early preg.	-	-14	-	-	Dieckmann and Wegner 1934 (21, p. 207)
36 postpart.	31	-14	34	rise	Thomson et al. 1938 (72, p. 51-55)
38.4 early preg.	36.1	-6	-	-	Roscoe and Donaldson 1946 (60, p. 532)
39.7 postpart.	37.7	-5	-	-	McLennan and Thouin 1948 (49)
38 postpart.	33.2	-12	36	rise	Tysoe and Lowenstein 1950 (73, p. 1195)
43 nonpreg.	38	-12	-	-	White 1950 (78)
36 nonpreg.	32	-11	-	-	Bucht 1951 (10, p. 45)
33 postpart.	30	-9	29-34	rise	Caton et al. 1951 (15, p. 1214-1215)
39 postpart.	31	-21	32	rise	Adams 1954 (2, p. 753)
36 postpart.	32	-11	19-24	rise	Werkö 1954 (77, p. 169-170
Average decrease:		11			

TABLE 8 HEMATOCRIT CHANGES ASSOCIATED WITH PREGNANCY (continued)

Control value (%)	Extreme value in preg. (%)	Max. change (%)	Time of occurrence of extreme	Occurrence of rise from extreme	Reference
COWS					
-	-	0	-	-	Feldman et al. 1936 (22)
33 postpart.	33	0	-	-	Reynolds 1953 (59)
SHEEP					
37 early preg.	28	-24	term	no rise	Barcroft et al. 1939 (3)
RABBITS					
39 prepreg.	32.5	-17	term	-	Zarrow and Zarrow 1953 (81)
-	-	decrease	term	-	Horger and Zarrow 1957 (37)
RATS					
50 prepreg.	36	-28	-	no rise	VanDonk et al. 1934 (74, p. 619)
-	-	-10	-	-	Bond 1948 (7)

the uterine vein. Burwell (14) also cited anatomical evidence for this view. That is, the structure of the maternal vascular system in the placenta offers a pathway between arteries and veins which would allow blood to flow through with little resistance, in much the same way that blood flows through a simple arteriovenous shunt.

Recently further evidence has been presented for this view by experiments of Glaviano (26). He found that clamping the uterine arteries of gravid dogs caused a decrease in cardiac output and a slight increase in blood pressure. This is similar to the changes resulting from the clamping of an arteriovenous fistula, but not the changes accompanying occlusion of arteries similar in size to those of the uterus, viz., femoral arteries.

A feature of the circulation in the pregnant woman unexplained by Burwell's theory is the reversal of the trend of cardiac output and blood volume a month or two before term. One present view is that these reversals are caused by an area of resistance growing within the shunt region, tending to block the flow of maternal blood across the placental site (McGaughey, 48). There is some anatomical evidence for this. Kline (43) reported that new tissue forms within the maternal intervillous spaces in the latter stages of pregnancy.

Uterine mass and vascularity increase considerably as

pregnancy proceeds and this increase in vascular space is enough to explain in part at least the circulatory changes of pregnancy. Estrogen is probably involved in the increased mass of the uterus in pregnancy. It has been shown to stimulate uterine growth and vascularity when administered to ovariectomized rats (Szego and Roberts, 71, p. 420-432). Urine levels of estrogen increase during pregnancy in all animals studied, such as man, macaque, cow, pig and rat (Zarrow, 80, p. 964). Zarrow suggests that this reflects an increased production and activity of this hormone.

Estrogen has been shown qualitatively to cause an increase in the quantity of blood flowing through the uterus when administered to ovariectomized rats (Holden, 36; Kalman, 42). Also, short term infusion of estrogen into nonpregnant ewes caused an increase in cardiac output and blood volume with no increase in arterial blood pressure (Parer, Metcalfe and Jones, 57). Estrogen has been shown to cause increased blood volume and decreased hemoglobin levels when injected into ovariectomized rats (Horger and Zarrow, 37). Also, estrogen injected into nonpregnant women resulted in increased blood volume and decreased hematocrit (Witten and Bradbury, 79).

Horger and Zarrow (37) investigated the interaction between estrogen and progesterone when injected into ovariectomized

rabbits. They found that at low levels of administered estrogen, progesterone caused an increase in the blood volume compared with the estrogen alone. However, at high levels of estrogen, progesterone partly inhibited the increase in blood volume resulting from estrogen alone. No anemia was noted in the rabbits treated with estrogen and progesterone in combination.

Brehm and Kindling (9, p. 705-708) suggested that the decreased peripheral resistance during pregnancy is due to the vessel dilating effect of progesterone. This decreased resistance then results in increased cardiac output and blood volume. However, no increase in cardiac output or decrease in peripheral resistance was noted when progesterone was injected into nonpregnant goats (Franklin, Herd and Metcalfe, 24) and sheep (Parer, Metcalfe and Moll, unpublished data). Furthermore, progesterone injected into ovariectomized rabbits did not cause any significant vascular changes (Horger and Zarrow, 37).

Newcomer (55) studied the relation between the hypophysis, ovary, placenta and fetus in the development of anemia of pregnancy in the rat. He found that in the presence of the placenta and the absence of the pituitary, ovary and fetus, anemia comparable to that in normal pregnancy occurred. He suggested that a hormone released by the placenta was responsible for the anemia of pregnancy

in the rat.

The role of hypervolemia and anemia in increasing cardiac output in pregnancy is uncertain. It has been suggested that increased blood volume, by increasing venous return to the heart, may increase the cardiac output. It is known that intravenous infusion of fluids (e.g. saline) increases the cardiac output (Huckabee, Casten and Harrison, 38). Fowler, Bloom and Ward (23) found that hypervolemia without anemia did not increase cardiac output of dogs, but hypervolemia with anemia or anemia without hypervolemia did increase it.

Hytten and Duncan (40) have discussed the role of anemia in the cardiodynamic changes in pregnancy. They suggest that the decreased hematocrit may result in a lowered resistance to blood flow because of the decreased blood viscosity. Hamilton (29) measured the relative blood viscosity in pregnant women and found that it was 3.8 at the sixth lunar month, compared with postpartum and nonpregnant control values of 4.6. However, the extent to which hypervolemia and anemia during pregnancy influence cardiodynamics has not been critically established.

Zarrow (80, p. 988-993) suggested that a multiplicity of factors seem to be involved in bringing about hypervolemia in pregnancy. He considers that sex steroids and other hormones are

important, and also the balances between them may bring about species differences in the maternal vascular adjustments to pregnancy.

III MATERIALS AND METHODS

Animals and Management

Nine white-face crossbred ewes of Columbia, Dorset Horn and Cheviot parentage were used. They were born between 1956 and 1959, and were three to six years old at the time of the experiment, in 1961-62. At the start of the experiment (November 1961) they were carrying approximately eight months of wool, and they were not shorn until after completion of measurements in July 1962, eight months later.

The sheep were housed in a barn with access to an open yard. They were communally fed pelleted alfalfa hay and concentrates, sufficient to gain weight throughout the experiment. The sheep were fasted from 24 to 30 hours before cardiovascular measurements were made. They were drenched at appropriate intervals to control parasites, and were apparently healthy throughout the experiment.

Before the experiment began the right carotid artery was exteriorized and placed in a tunnel of skin to facilitate catheterization (Bone, Metcalfe and Parer, 8). Adequate time for healing of the site was allowed before cardiovascular measurements began.

Five of the sheep were mated to a Southdown ram in November-December of 1961. The brisket of the ram was smeared with paint, and the day of mating was determined from markings on the backs of the ewes. Four nonpregnant sheep served as controls.

The pregnant sheep delivered in April-May. Lambs were removed at birth to eliminate or reduce the effects of lactation. Details of breeding and delivery are given in IV Results.

Explanation of the Techniques Used

Cardiac Output. Cardiac output is defined as the volume of blood ejected by one ventricle of the heart per minute. Ideally it would be measured by a flowmeter placed at the root of the pulmonary artery. However, in the absence of such equipment, it can be measured indirectly using the Stewart-Hamilton method (Stewart, 68; Hamilton, Moore, Kinsman and Spurling, 31). This is based on the principle that if an indicator substance is injected into a flowing liquid, the degree of dilution (after adequate mixing) is a measure of the volume of flow. Using this technique, one follows the passage of a dye through the heart. For a given quantity of dye injected into a vein, the cardiac output will be a function of the average concentration of the dye as it issues from the heart (the dye having mixed completely with the blood within the heart), and the time of one passage of the dye through the heart, i.e.,

$$(1) \qquad F = \frac{I \times 60}{c \times t}$$

where F = flow rate per minute,

I = amount of dye injected,

c = average concentration of dye after passage through the heart,

t = time in seconds of one passage of all the dye through the heart.

The rationale of this principle can be seen as follows: consider water flowing from a pipe at a constant rate,

$$(2)\ \text{Flow per min.} = \frac{V \times 60}{t}$$

where V = the volume flowing out of the pipe in time interval t in seconds (Fig. 1).

If the volume cannot be measured, but the flow can be stopped, a known amount of indicator dye could be mixed with the water and the concentration of the dye in the water determined (Fig. 2). Then,

$$(3)\ \text{Volume of water} = \frac{I}{c}$$

where I = amount of indicator,

c = concentration of the indicator after mixing.

Substituting equation (3) in equation (2), the flow rate becomes

$$(1) \qquad F = \frac{I \times 60}{c \times t}.$$

Considering the pipe again, suppose that the flow cannot be stopped, but the dye can be injected into the pipe, and the dye-water mixture can be sampled at a point below the point of the injection (Fig. 3).

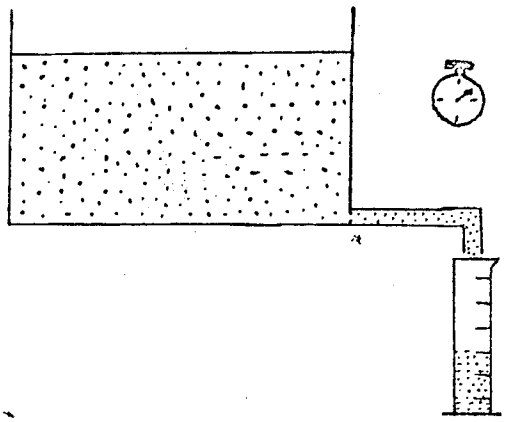


Fig. 1 Flow rate measured by collecting and measuring outflow over a timed interval.

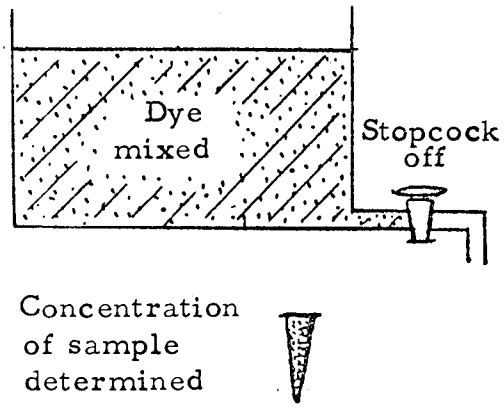


Fig. 2 Volume in reservoir measured by adding a known quantity of an indicator dye and measuring its concentration after mixing.

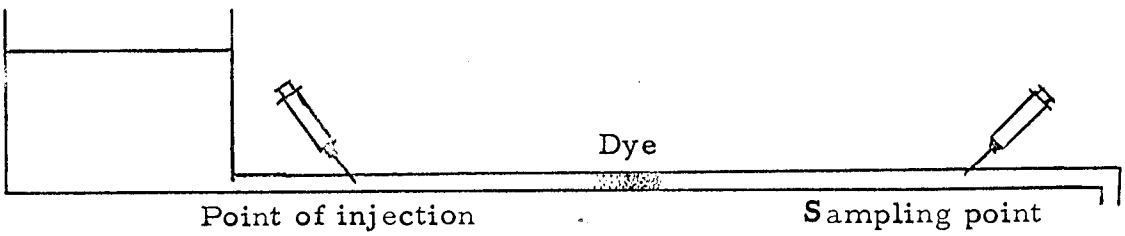


Fig. 3 Flow rate through this pipe is measured by injecting a dye, withdrawing at a constant rate from the sampling syringe during passage of the dye, and measuring the mean concentration of the dye in the sampled liquid, and its time of passage past the sampling point. (After Rushmer, 62, p. 88)

The sampling point must be such that the dye has mixed completely with the liquid passing any given point in the pipe. Now the mean concentration of the dye in the water passing the sampling point will be the same as the mean concentration in a continuously collected sample of the liquid as it passes the sampling point. The time taken for the sampled liquid (with its dye) to pass the sampling point will be the same as the time taken to fill a volume V at a concentration c (V being the volume of liquid with which the dye has mixed, and c being the mean concentration of the dye in the liquid passing the sampling point).

This discussion can be applied to the blood circulatory system. A known quantity of dye can be injected into a vein, and the blood containing the dye, after passing through the heart and mixing completely with the blood present in the heart, can be sampled from an artery (Fig. 4). The concentration of dye in the blood can be measured by passing the sampled blood (being withdrawn at a constant rate) through a cuvette with a light source on one side and a photosensitive element on the other side. The varying density of the dye-blood mixture will cause a variation in the amount of light passing through the blood-filled chamber. This will change the electrical signal from the photosensitive element, which in turn can be amplified, and recorded. The characteristics of the instrument can

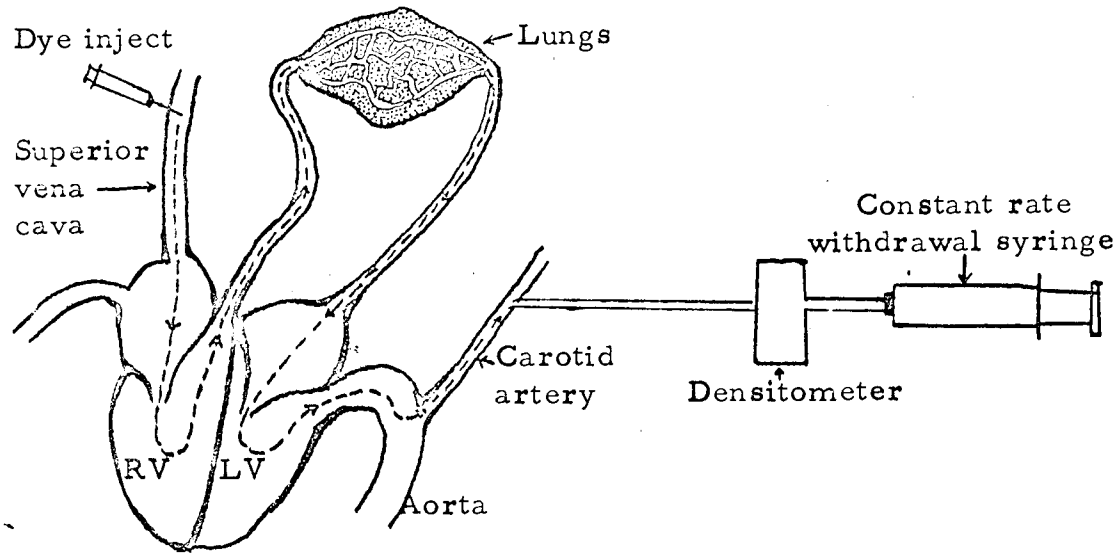


Fig. 4 Schematic diagram showing passage of the dye through the vena cava, heart and pulmonary vessels, and thence to the carotid artery from where it is sampled.

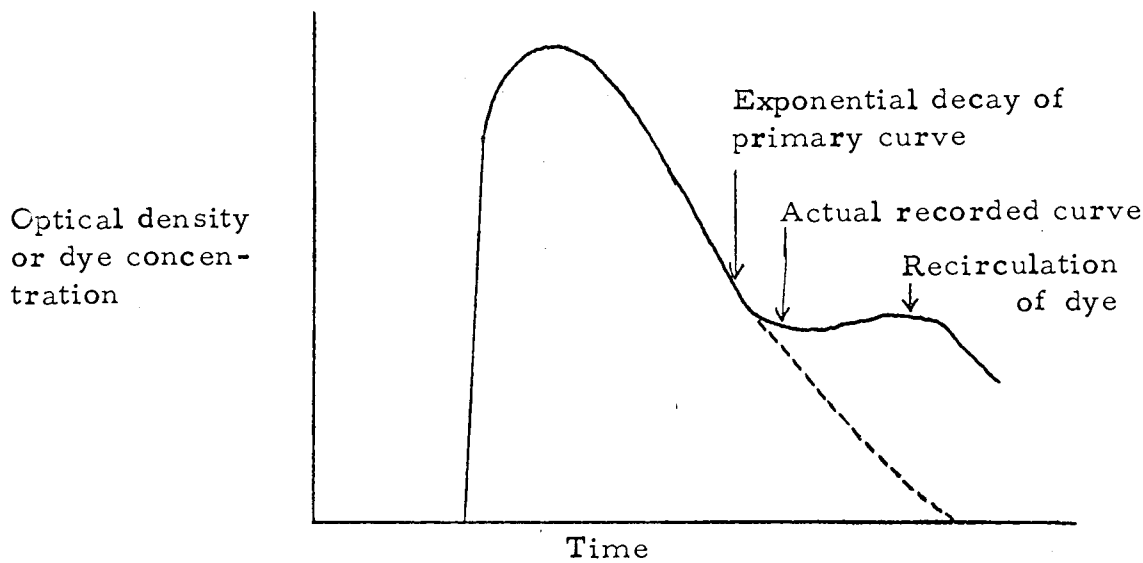


Fig. 5 Optical density (or dye concentration) curve obtained after injection of dye into a vein, and continuous sampling of blood from an artery during the first and second circulation of the dye through the heart. The dotted curve is the completion of the theoretical primary circulation of the dye.

be such that the output of the machine is directly proportional to the optical density and, therefore, the dye concentration of the sampled blood.

The characteristic curve obtained is one in which the dye concentration suddenly rises, and then falls (Fig. 5). This decay is exponential, but before it has completed its full course, recirculation of the dye occurs. The reason for this can be seen in Figure 6. The central pump represents the heart. Blood flowing through the shorter circuits carries dye back to the heart before all of the original injected bolus of dye has been washed out of the heart.

As seen from equation (1), we must know the full curve of primary passage of the dye through the heart in order to estimate the average concentration of the dye in its passage past the sampling point. In fact, the primary curve can be calculated because the exponential decay begins before recirculation occurs. This decay, plotted on semilogarithmic paper, gives a straight line. With appropriate calibration (i. e. knowing the deflection produced by a given dye concentration) the curve can be used to measure mean concentration of the dye, and the time of one passage through the heart.

Many methodological refinements have been made in the dye-dilution method for the measurement of cardiac output. The method used in the present work will be briefly described.

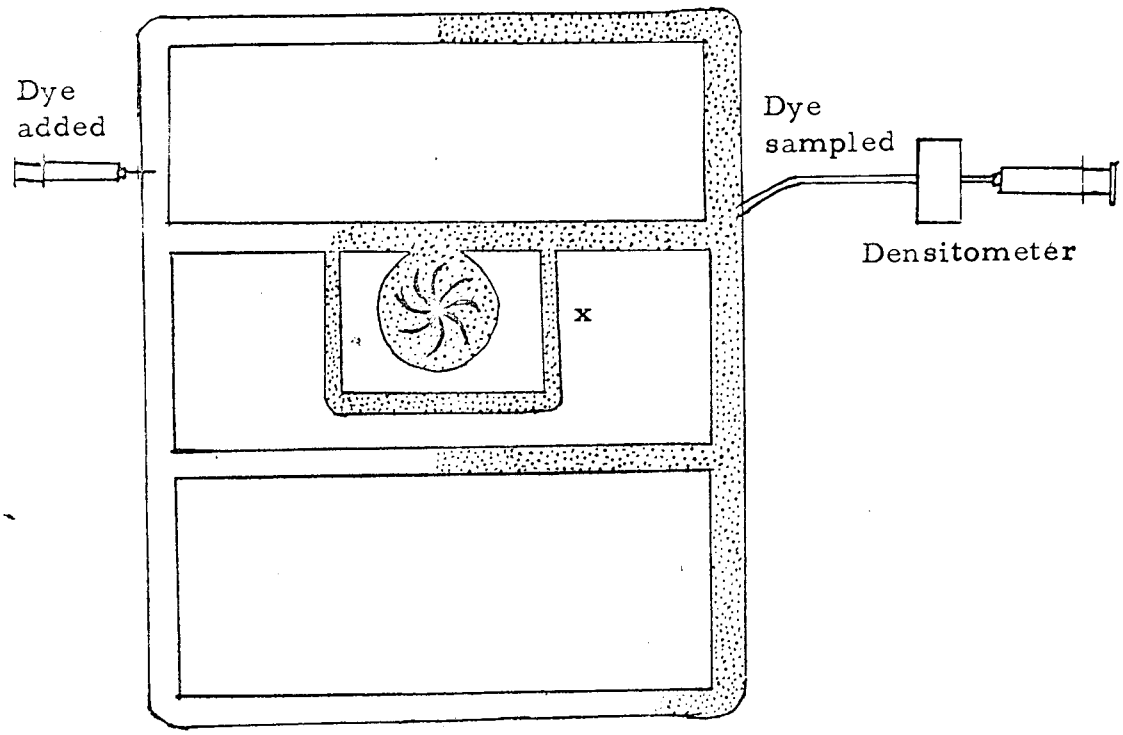


Fig. 6 Hydraulic model showing that some of the injected dye can return to the heart through some vessels (x) before the completion of the first passage of the dye through the heart. (After Rushmer, 62)

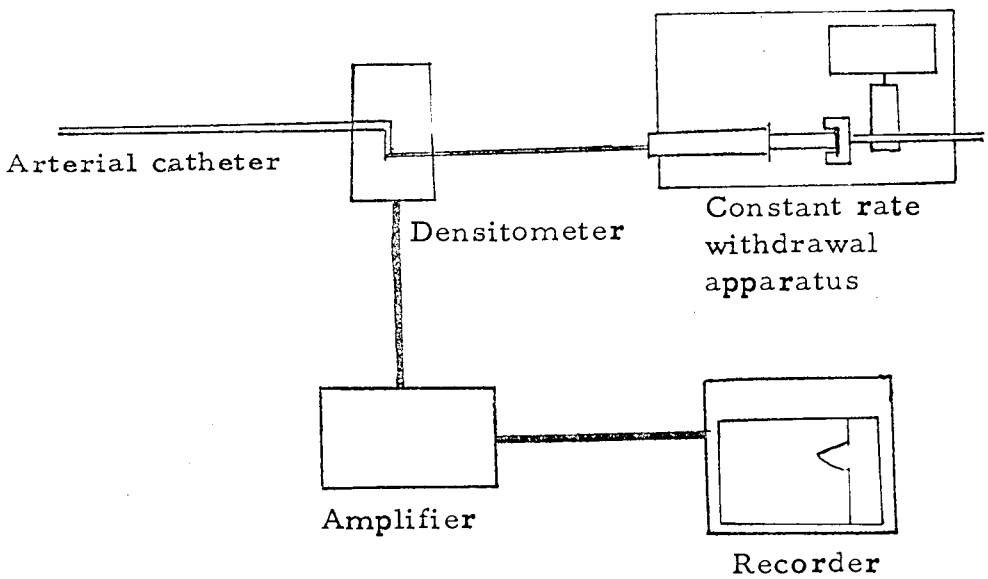


Fig. 7 Schematic diagram of apparatus used for obtaining dye dilution curves for calculation of cardiac output.

Dye was injected into the jugular vein and the blood sampled from the carotid artery (Fig. 4). The blood was taken from the carotid artery using a constant rate withdrawal apparatus, the blood being withdrawn through a continuously recording densitometer (Fig. 7). The signal produced by the variation in dye concentration was amplified and recorded. The blood collected during the sampling period (pooled sample) was mixed thoroughly, and the optical density of dye in the plasma determined with a spectrophotometer. This was read against a plasma blank collected immediately before injection of the dye. The blood sampling was discontinued when it was judged that the exponential curve had been recorded, but before recirculation had commenced. It is not essential that sampling of blood should cease before recirculation of the dye, though calculations are simplified if it does. Figure 8 shows an idealized but typical curve which was obtained.

In order to see how the cardiac output is calculated from this method, let us return to the original equation,

$$(1) \qquad F = \frac{I \times 60}{c \times t}.$$

Instead of measuring the actual amount of dye injected into the animal, we can inject an equal quantity I into the animal, and also into a volumetric flask.

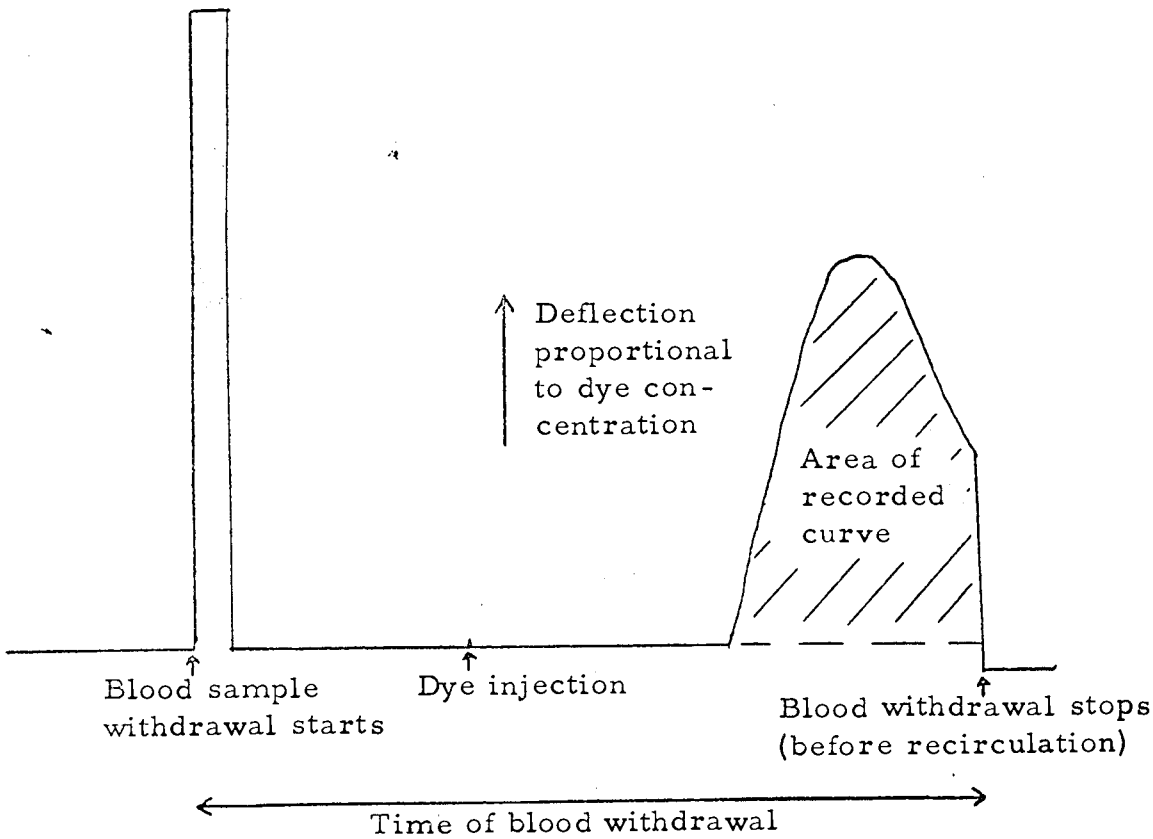


Fig. 8 A typical recorded curve showing its important characteristics.

Now, I = volume x concentration.

The optical density of the standard can be measured by using a spectrophotometer. Optical density is directly proportional to concentration, the proportionality factor being K.

Thus, I = volume of standard (V_{std}) x optical density (OD_{std}) x K,

so $$F = \frac{V_{std} \times OD_{std} \times K \times 60}{c \times t}.$$

Now the average concentration in the primary curve, c =

$$\text{average deflection in the primary curve} \times \frac{\text{concentration of dye}}{\text{unit deflection}}.$$

$$\text{Average deflection in the primary curve} = \frac{\text{area of primary curve}}{\text{time of primary curve}}.$$

Concentration of dye/unit deflection =

$$\frac{\text{concentration of dye in plasma of pooled sample}}{\frac{\text{area under recorded curve}}{\text{time of blood withdrawal}}} =$$

$$\frac{\text{optical density of pooled sample} \times K}{\frac{\text{area under recorded curve}}{\text{time of blood withdrawal}}}.$$

So flow rate =

$$\frac{V_{std} \times OD_{std} \times K \times 60}{\text{time of primary curve} \times \frac{\text{area of primary curve}}{\text{time of primary curve}} \times \frac{OD \text{ sample} \times K \times \text{time of blood withdrawal}}{\text{area under recorded curve}}} =$$

$$\frac{V_{std} \times OD_{std} \times 60 \times \text{area under recorded curve}}{\text{area of primary curve} \times OD_{sample} \times \text{time of blood withdrawal}}.$$

The above is plasma flow rate because the dye is distributed only in the plasma and not in the cells.

$$\text{Blood flow rate} = \frac{\text{plasma flow rate}}{(1 - \text{hematocrit})}.$$

Hematocrit is readily estimated as the percent packed cell volume.

Blood Volume. The plasma volume was measured by the dilution technique using Evans blue dye (T. 1824). A known amount of the dye is injected into the animal, and the same quantity of dye is injected into a two-liter volumetric flask of distilled water. Ten minutes after injection of the dye (i.e. when the dye has mixed with the blood), a blood sample is taken from the animal. After centrifugation, the plasma is removed and the optical density of the Evans blue in the plasma is measured in a spectrophotometer (at 620 mμ) against a reference of plasma collected immediately before injection of the dye.

$$\text{Plasma volume} = \frac{\text{amount of dye injected}}{\text{concentration of dye in plasma after mixing}}.$$

Amount of dye = amount of dye injected into standard flask =

$Vol_{std} \times \text{optical density}_{std} \times K$ (a constant).

Concentration of dye in plasma = $OD_{sample} \times K$.

$$\text{Plasma volume} = \frac{Vol_{std} \times OD_{std} \times K}{OD_{sample} \times K}.$$

$$\text{Blood volume} = \frac{V_{std} \times OD_{std}}{OD_{sample} \times (1 - \text{hematocrit})}.$$

The validity of the Evans blue method has been demonstrated in pregnant cattle by Reynolds (59), and there is no reason to believe it would not also be valid for pregnant sheep.

Arterial Blood Pressure and Pulse Rate. In this work the arterial blood pressure was measured by attaching a resistance wire strain gauge transducer directly to the arterial catheter. In this instrument variations in blood pressure work on the plunger of the transducer, resulting in variations in the degree of stretching of wires which alter their resistance to electrical current. This causes alterations to the balance of a Wheatstone bridge, in proportion to the applied pressure, and the resulting output can be amplified and recorded (Rushmer, 62, p. 140-141).

The recorded tracing must be calibrated, and this is done using a mercury manometer. Known pressures are applied to the transducer (e.g. 50 and 100 mm Hg) and the deflection recorded on the paper. The systolic and diastolic arterial pressures can then be calculated from the tracing. By knowing the speed of the recording paper, the number of heat beats per minute can also be counted.

Experimental Procedure and Equipment

Measurements were made on each sheep at approximately

three-weekly intervals. On each experimental day measurements could be made on up to four different ewes. Each sheep was transported the short distance from the barn to the laboratory, and weighed to the nearest pound. It was then placed in a loose canvas sling suspended within a wooden stanchion. The floor of the stanchion was approximately 15 inches above floor level. The sheep was not completely suspended in the sling, as its purpose was only to prevent the sheep moving away when catheters and equipment were in place. The air temperature of the laboratory was approximately 21° C, and each sheep was in the room for about 1-1/2 hours before all measurements were completed (Fig. 9).

The wool was shorn from the area around the carotid loop and on the opposite side over the jugular vein (Fig. 10) using fine clippers. A disinfectant (pHisoHex) was applied to the skin over the carotid loop and the jugular vein. The carotid loop was then injected subcutaneously with 2-3 cc of a local anesthetic, Xylocaine. A small incision was made in the skin above the artery with a scalpel and an 18-gauge thin-wall hypodermic needle was inserted into the carotid artery directed towards the heart. Blood flow from the needle was prevented by clamping the loop just below the needle point. A leader of nylon fishing line was placed through the needle into the artery and the needle removed. After wiping the exposed part of the leader

Fig. 9 A sheep in the stanchion awaiting cardiovascular measurements.

Fig. 10 Neck of a ewe showing the carotid loop. The gauze is placed behind the loop only for illustrative purposes.

free of blood, a catheter could be threaded over the leader until it was well inside the artery. The arterial catheter used was 50 cm of Intramedic PE 190 polyethylene tubing (outside diameter 0.067 in, inside diameter 0.047 in). The catheter was prepared by drawing out one end until it fitted snugly over the leader. This allowed easy access of the catheter through the skin and artery wall. The other end was fitted to a Luer-Lok adapter, using a flared end of the catheter as a gasket. After removal of the leader from the artery, the catheter was filled with physiological saline containing 1% by volume of heparin solution (1000 units/cc).

The site of the jugular vein puncture was anesthetized with Xylocaine, the skin incised and a six-gauge hypodermic needle placed into the lumen of the vein. A catheter of 50 cm of polyvinyl plastic tubing (outside diameter 0.070 in, inside diameter 0.038 in) was inserted through the needle into the vein. The catheter was filled with heparinized saline and taped into place.

Arterial blood pressure was then measured, using a New Electronic Products resistance wire strain gauge transducer and amplifier. The strain gauge was placed on a fixed reference bench a few cm above heart level, the error involved depending on the size of the sheep. Thus the absolute values of pressure may be a few units (mm Hg) low, but serial measurements on individual sheep

would give valid comparisons for detecting changes during pregnancy. The arterial pressure was recorded on a Texas Servo/Riter recorder. After 15-30 seconds of recording, the transducer was detached from the catheter, and a zero (atmospheric pressure) level was recorded. The transducer was then calibrated with a mercury manometer (using a modified sphygmomanometer) and intervals representing known pressure differences were recorded on the paper.

The cardiac output was next measured. Two operators were needed for this; one to inject the dye, and the other to operate the densitometer and ancillary equipment.

The dye injection apparatus consisted of two syringes connected to an open T-piece (the latter was a modified three-way stopcock). The third arm of the T-piece was connected to the venous catheter. One syringe had a stop so that when the syringe was filled it always contained a constant volume of dye, approximately 1 cc of 0.5% Evans blue being used. The second syringe was mounted behind a one-way valve, and it contained 5 cc of physiological saline for flushing the dye from the catheter. As explained in III Materials and Methods (Cardiac Output), the important feature was not the quantity of dye that the syringe contained, but only that it deliver the same quantity to the animal and to the volumetric standard flask. Duplicate standards invariably showed good agreement between successive

injections from the apparatus.

The dye injection operator attached the syringe to the venous catheter, while the densitometer operator cleared the arterial catheter of saline and drew a sample of blood (approximately 7 cc) to be used as a blank for the readings of optical density of dye in the later samples. Then he attached the arterial catheter to the densitometer cuvette intake and turned on the constant withdrawal apparatus.

A sharp increase in optical density marked the time of commencement of blood withdrawal. A baseline level of density of blood was recorded during this period (Fig. 11). When the baseline was steady the dye was injected. A few seconds later the optical density rose sharply (as seen on the recorded curve), followed by the characteristic exponential washout (Fig. 8). When a good portion of the decay was recorded, but before recirculation had occurred, a stopcock was turned so that air was drawn through the densitometer (Fig. 12). A rapid decrease in optical density marked the end of blood withdrawal. The blood which had been collected over this withdrawal period (generally about 10-15 cc) was mixed and a sample taken for centrifugation. The unused blood was returned to the sheep. The optical density of Evans blue dye in the plasma read against the blank collected before injection is that referred to as "O. D. (optical

Fig. 11 Measurement of cardiac output. The operator on the left is about to inject dye into the jugular vein catheter, while the one on the right is supervising equipment for measurement of concentration of the dye in continuously sampled arterial blood.

Fig. 12 At the end of the operation, blood sampling from the artery is discontinued and the dye curve has been drawn on the recorder in the foreground.

density) of pooled sample" in III Materials and Methods (Cardiac Output).

The arterial catheter was cleared of blood and filled with heparinized saline. Ten minutes after the dye injection a second determination of cardiac output was carried out as described, and in another ten minutes the process was again repeated. The blanks taken ten minutes after each injection served for the determination of plasma volume (III Materials and Methods, Blood Volume). A final blood sample was taken ten minutes after the last injection, thus giving three determinations of plasma volume.

The blood samples were centrifuged until the hematocrit remained constant. This was tested on the machines used (clinical centrifuges), and it was found that 50 minutes at a certain speed was sufficient. Seven cc blood samples were taken, and centrifugation was carried out in graduated centrifuge tubes so that hematocrit could be readily calculated from the proportion of the packed cells. The plasma was transferred to matched cuvettes (13 mm diameter) and its optical density read on a Coleman Junior Spectrophotometer at a wavelength of 620 mμ.

The sheep was given intravenously one cc of iron dextran (Proferrin, 20 mg available iron per cc) after the final sample of blood was taken, so that there would be no difficulties with iron

defiency due to the repeated removal of blood from the sheep. Less than 100 cc, or approximately 2% of the blood volume, was taken during the total procedure. The catheters were removed from the vein and artery, slight pressure being applied to the artery until bleeding from the incision stopped. pHisoHex was applied over the punctures and the sheep was released.

The Evans blue standard was made by mixing the same quantity of dye that was injected into the sheep in a two liter volumetric flask with distilled water. The optical density of this was read against a distilled water blank at a wavelength of 620 mμ on the spectrophotometer.

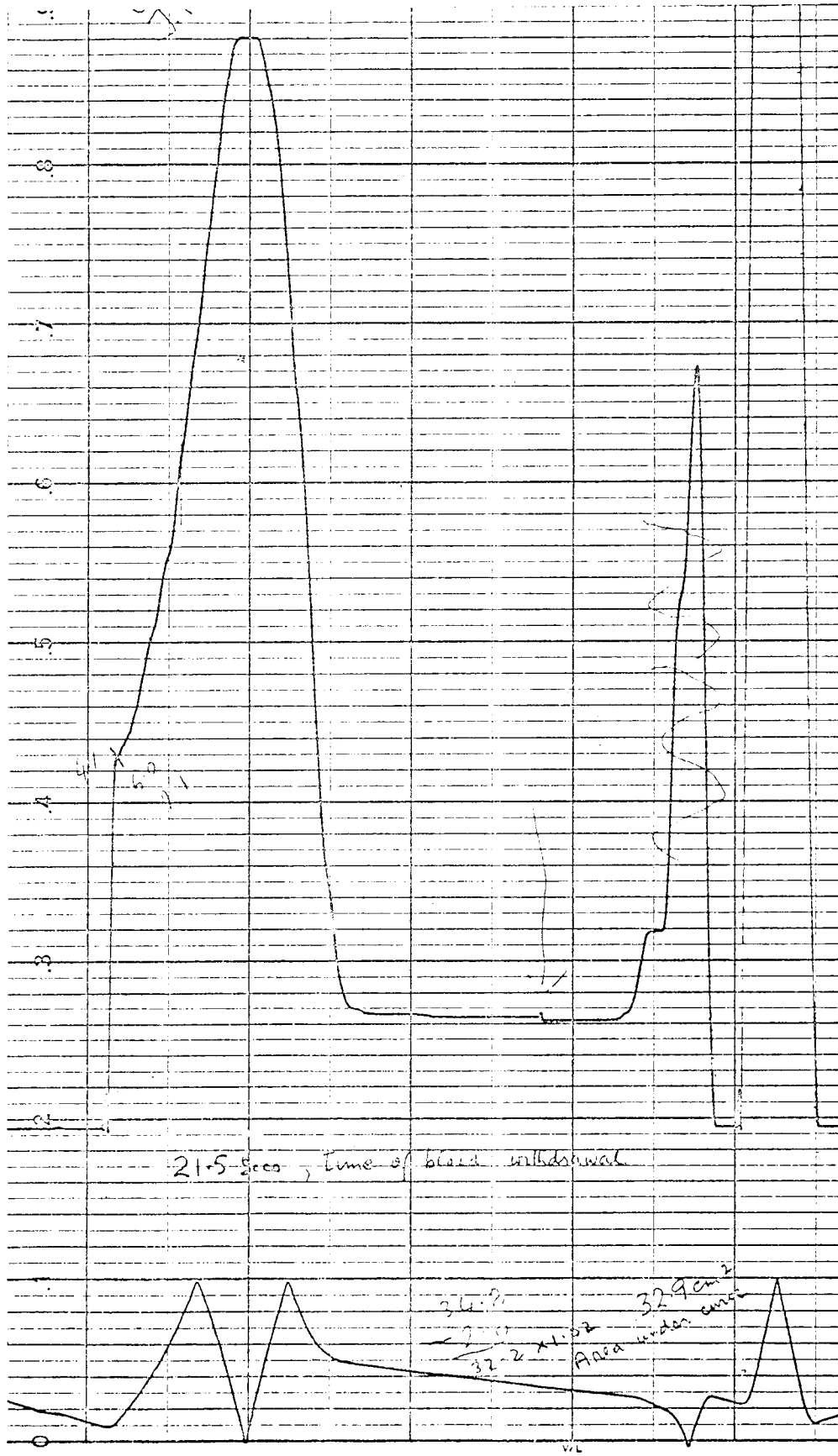
The equation for cardiac output (derived in III Materials and Methods, Cardiac Output) is as follows: cardiac output =

$$\frac{Vol_{std} \times OD_{std} \times 60 \times \text{area under recorded curve}}{\text{area of primary curve} \times OD_{sample} \times \text{time of blood withdrawal} \times (1 - hct)}$$

All of these values can be obtained from the recorded tracing and the spectrophotometer. The area under the curve can be measured by a planimeter, or obtained directly by an integrator attached to the recorder which gives the area in cm^2. The area of the theoretical primary curve can be estimated by adding the area of the recorded curve before recirculation to the remainder of the

exponential decay, which can be calculated when the decay characteristic is known. An actual curve and calculation sheet are shown in Figures 13 and 14.

Fig. 13 An actual recording of a dye dilution curve, drawn from right to left. Important features of the curve are shown in Figure 8. The lower tracing is that of the integrator, which allows easy calculation of the area under the curve. Calculation of the cardiac output is shown in Figure 14.


21·5 secs
Time of blood withdrawal
32·9 cm²
Area under curve

Subject: Sheep G137
Date: May 25, 1962
C. O. #: 1
Remarks: Subject was standing quietly

CALCULATION OF CARDIAC OUTPUT AND BLOOD VOLUME

I. Data from recorded tracing
   A. Area under recorded curve (cm^2) 32.9
   B. Time of blood withdrawal (sec) 21.5
   C. Area under recorded curve before recirculation 32.9

II. Data from semilog replot of primary circulation curve
   A. T/2 = time (in cm) for concentration to decline 50% 0.85
   B. Cb = concentration (in cm) from baseline at end of recording (before recirculation) 4.10
   C. Area under extrapolated curve = 1.44 x T/2 x Cb = 1.44 x .85 x 4.10 = 5.02

III. Area under primary curve = I. C. + II. C. = 32.9 + 5.02 = 37.92

IV. Other data
   A. O. D. standard .232
   B. Volume standard 2 Liters
   C. O. D. pooled plasma .228
   D. Hematocrit 20/73 27%
   E. Weight of subject 140 lbs 63.6 kg

V. Cardiac Output Calculation

$$C.O. = \frac{\text{vol. stand. (L)} \times \text{O. D. stand.} \times \text{area under recorded curve} \times 60}{\text{area under primary curve} \times \text{O. D. pooled plasma} \times \text{time of withdrawal (sec)} \times (1 - \text{hct})}$$

$$\frac{2 \times .232 \times 32.9 \times 60}{37.92 \times .228 \times 21.5 \times .73} = 6.75 \text{ L/min}$$

$$\frac{\text{C.O. (L/min)}}{\text{Wt of subject (kg)}} = \frac{6.75}{63.6} = 106 \text{ ml/kg/min}$$

$$\text{Plasma volume} = \frac{\text{vol. stand.(L)} \times \text{O. D. stand.}}{\text{O. D. of 10 min. sample}} = \frac{2 \times .232}{.138} = 3.36 \text{ L}$$

$$\text{Blood volume} = \frac{\text{plasma vol.}}{(1 - \text{hct})} = \frac{3.36}{.73} = 4.61 \text{ L}$$

$$\frac{\text{Blood vol.}}{\text{Wt of subject (kg)}} = \frac{4.61}{63.6} = 73 \text{ ml/kg.}$$

Fig. 14 Calculation sheet for cardiac output and blood volume.

IV RESULTS

The results are presented in both tabular and graphic form. Individual values for each sheep on each experimental day are presented in the graphs. The data in the tables are average values for each group of sheep (pregnant and nonpregnant) at various time intervals before and after parturition.

Statistical Analyses of the Results

Two major analyses of the data were carried out. In the first analysis (Analysis A) the regression coefficient (i. e. slope of the regression line) for each measured function of each sheep for the 90 or 150 days (if the data were available) before delivery was calculated. The regression coefficients for the same functions were calculated over approximately the same chronological period in the nonpregnant ewes. These two sets of regression coefficients were then analyzed by the t-test.

In the second analysis (Analysis B) the mean value of each function for each sheep for 30 days after delivery was subtracted from the mean value for the same sheep for the last 30 days of pregnancy. The differences were then compared with those similarly obtained in the four nonpregnant ewes by means of the t-test.

The variability of the triplicate measurements of cardiac

output, blood volume and hematocrit carried out on each sheep on each measurement day was estimated. The mean deviation of each measurement from the mean of the three determinations was calculated and expressed as a percentage of the mean.

Breeding and Delivery of the Ewes

Table 9 gives details of the breeding date, gestation length, size and sex of the lambs produced.

The mean delivery date was April 29, 1962, and this was the date used as day 0 in constructing the graphs of functions of the nonpregnant sheep (see below). The mean recorded gestation length was 146 days, the standard deviation being 3.9 days. Both the length of gestation and the birth weights of lambs were within normal limits, though two sheep produced dead offspring. The twins produced by H 300 showed no signs of decomposition, while the lamb of I 46 showed signs of decomposition that indicated death had occurred in utero approximately one week prior to delivery (as estimated by a veterinarian) though birth weight was normal.

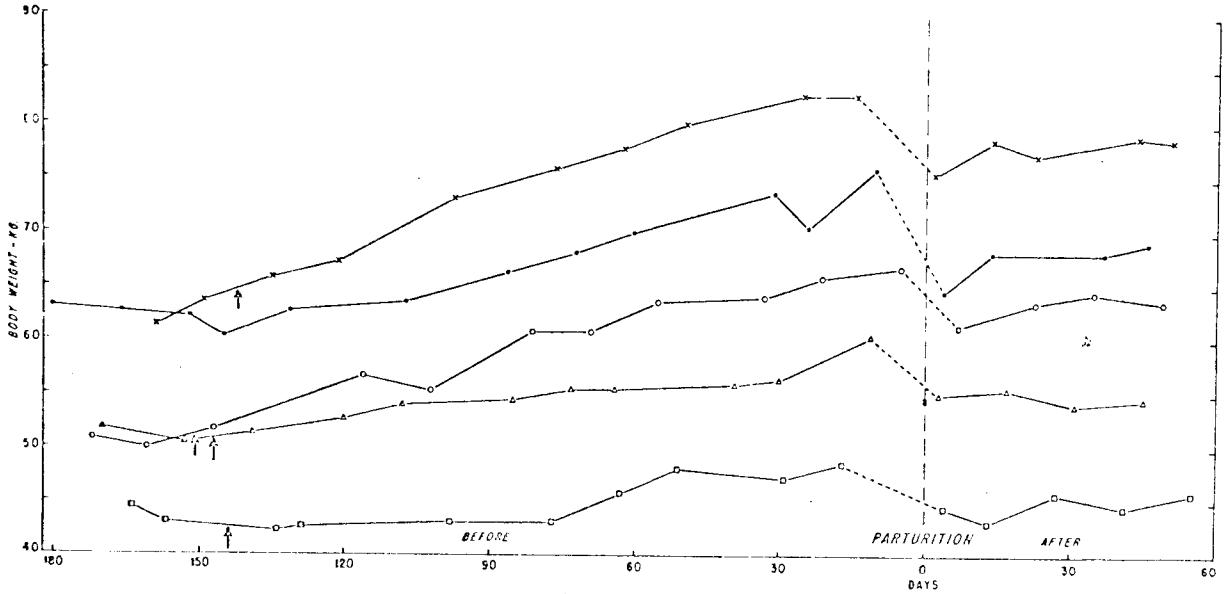
Body Weight

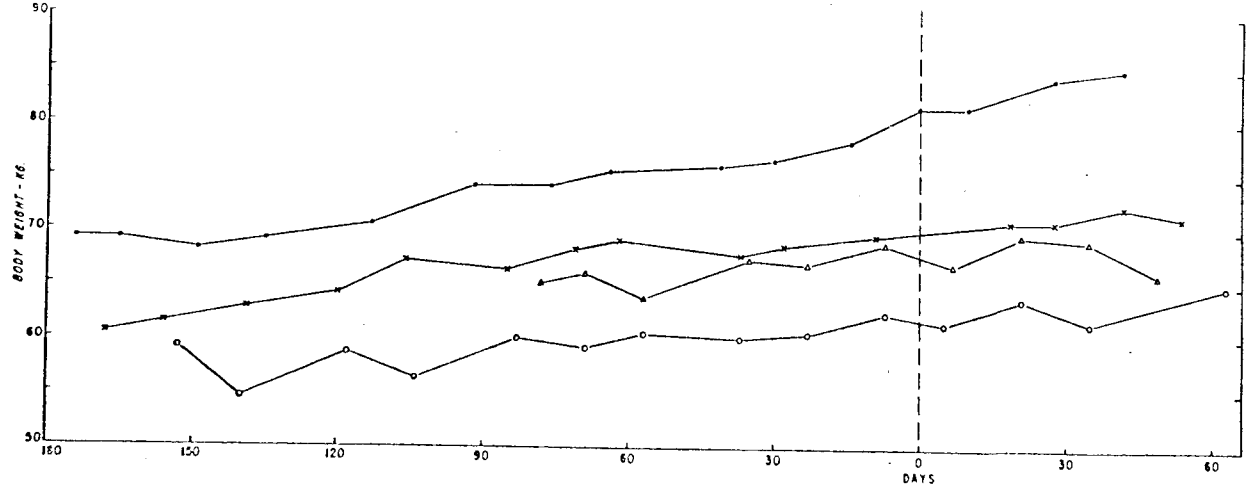
Body weights of all animals are shown in Figure 15. The upper graph represents the pregnant sheep, and the lower graph the nonpregnant sheep. Arrows mark the known conception dates. Group

TABLE 9 BREEDING DATA OF THE SHEEP

Sheep No.	Breeding date (1961)	Delivery date (1962)	Gestation (days)	Lambs		
				Weight (kg)	Sex	Remarks
H300	Nov. 27	Apr. 18	142	-	FF	twins, stillborn
H 62	Dec. 5	Apr. 28	144	4.5	M	-
I 46	-	Apr. 30	-	3.2	-	dead approximately one week
G137	Dec. 2	May 2	151	3.2	F	-
G 11	Dec. 10	May 6	147	4.5	F	-

Fig. 15 Body weights of the pregnant sheep (upper graph) and the nonpregnant control sheep (lower graph) during approximately the same chronological period. Arrows represent known dates of mating.


BODY WEIGHT - KG
90
80
70
60
50
40
180
150
120
90
60
30
0
30
60
DAYS
BEFORE
PARTURITION
AFTER


BODY WEIGHT - KG
90
80
70
60
50
180
150
120
90
60
30
0
30
60
DAYS

averages at three week intervals are given in Table 10. The point marked "0" in the lower graph (Fig. 15), i. e. the graph of body weights of the nonpregnant ewes, is the same as the average delivery date of the pregnant ewes. The body weights of both groups increased during the experimental period, the nonpregnant group by an average of 8 kg, and the pregnant sheep by an average of 13 kg at term. There was then a decrease of 5 kg after delivery in the pregnant sheep. The difference between pregnant and nonpregnant groups in change of body weight was statistically significant according to Analysis B ($P < 0.01$), when 30-day postpartum values were subtracted from 30-day prepartum values.

Cardiac Output

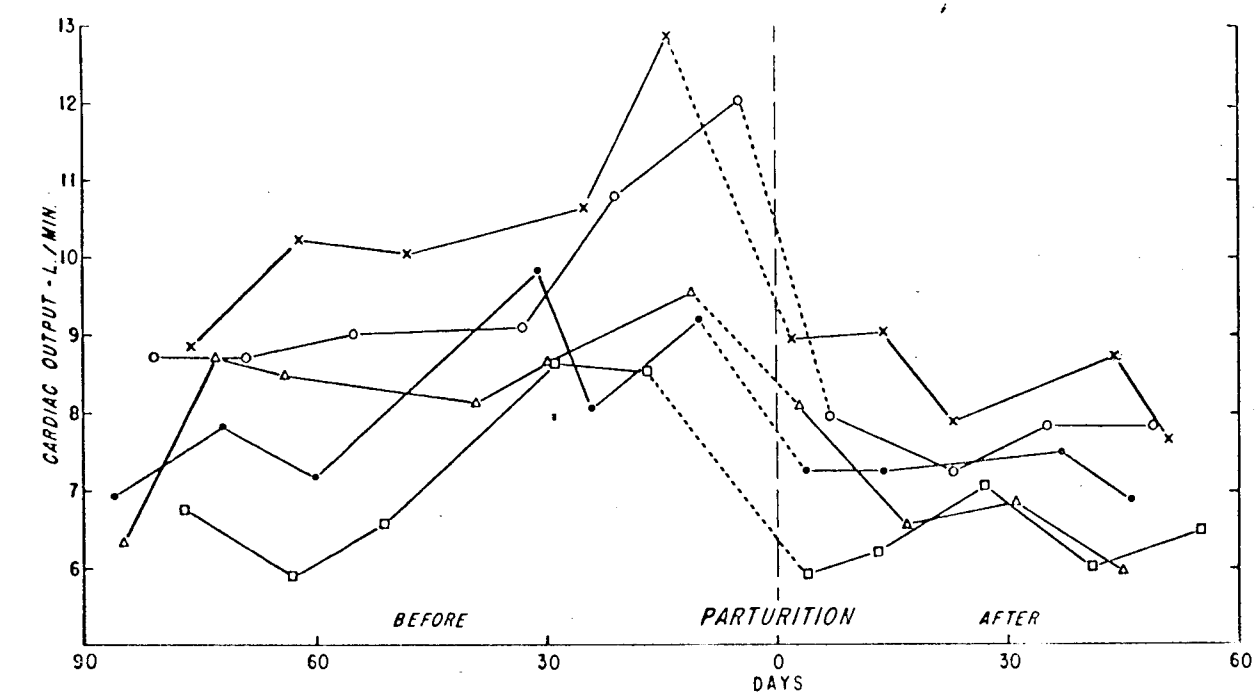
The cardiac output changes associated with pregnancy are shown in Figure 16 and Table 11. Measurements of cardiac output were made commencing at about the tenth week of pregnancy. Probably at this stage there was already some increase in cardiac output above control levels. In the last three weeks of pregnancy the cardiac output of the pregnant ewes averaged 10. 3 liters per minute, this being 41% greater than the average postpartum (0-9 weeks after delivery) value. Compared to this increase the changes occurring in the nonpregnant ewes during the same period were relatively minor. According to both statistical analyses, the differences

TABLE 10 AVERAGE BODY WEIGHT OF PREGNANT AND NON-PREGNANT EWES

Time (weeks)	Body weight (kg)	
	Pregnant*	Nonpregnant+
Before pregnancy		
3 - 0	54	63
During pregnancy		
0 - 3	55	62
4 - 6	57	65
7 - 9	59	66
10 - 12	61	67
13 - 15	63	67
16 - 18	65	68
19 - 21	67	70
Postpartum		
0 - 9	62	71
Increase above postpartum at term:	8%	-1%
Significance level (P): Analysis A	< 0.1	
Significance level (P): Analysis B	< 0.01	

*Five pregnant ewes.
+Values for four nonpregnant ewes during the period when the other five were pregnant.

Fig. 16 Cardiac output of the pregnant sheep (upper graph) and the nonpregnant control sheep (lower graph) during approximately the same chronological period.


CARDIAC OUTPUT - L./MIN.
13
12
11
10
9
8
7
6
BEFORE
PARTURITION
AFTER
90
60
30
0
30
60
DAYS

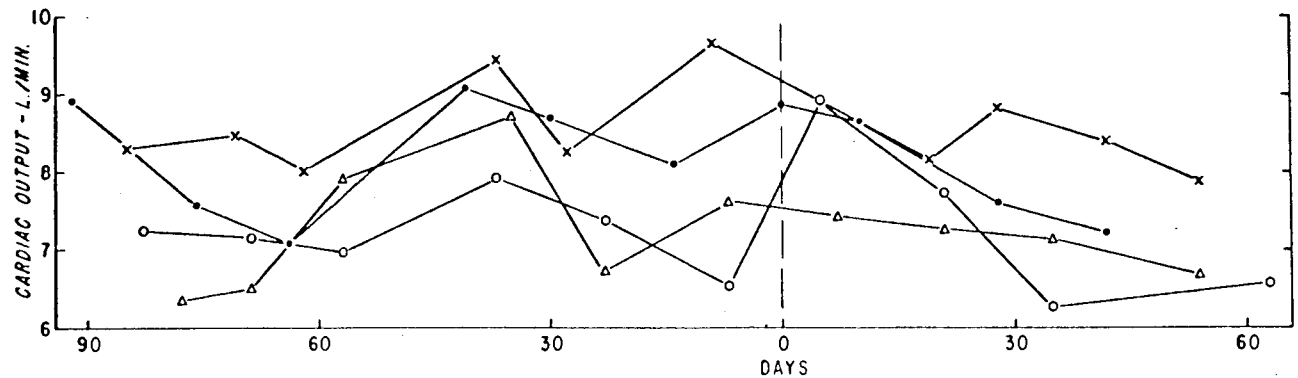

CARDIAC OUTPUT - L./MIN.
10
9
8
7
6
90
60
30
0
30
60
DAYS

TABLE 11 AVERAGE CARDIAC OUTPUT OF PREGNANT AND NONPREGNANT EWES

Time (weeks)	Cardiac output (liters/min)		(ml/kg/min)	
	preg.*	nonpreg.+	preg.*	nonpreg.+
During pregnancy				
10 - 12	8.0	7.5	134	111
13 - 15	8.3	7.5	132	112
16 - 18	9.2	8.3	146	125
19 - 21	10.3	8.1	157	115
Postpartum				
0 - 9	7.3	7.5	120	106
Increase above postpartem at term:	41%	8%	31%	8%
Significance level (P):				
Analysis A	<0.01		<0.01	
Analysis B	<0.01		<0.02	

*Five pregnant ewes.
+Values for four nonpregnant ewes during the period when the other five were pregnant.

between pregnant and nonpregnant sheep were significant ($P < 0.01$).

Figure 17 and Table 11 contain the cardiac output results, expressed as the cardiac output per unit of body weight. This increased to a value of 157 ml/kg/min at term, 31% above the postpartum control value of 120 ml/kg/min. The change was statistically significant (A: $P < 0.01$ and B: $P < 0.02$). The values in the nonpregnant sheep showed relatively minor fluctuations.

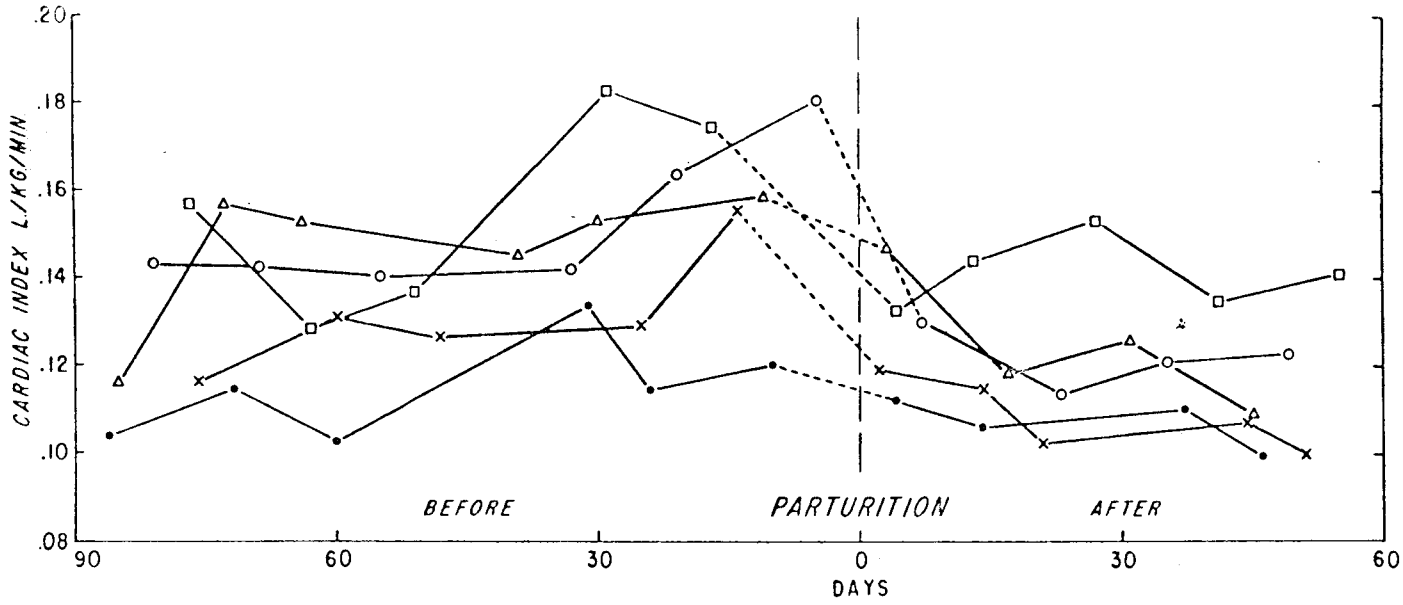
The mean percent deviation of single cardiac output measurements from the mean of the three done on that day averaged 5.9% overall. This includes ten days when triplicate measurements were made on each of the nine sheep, or 270 individual determinations of cardiac output altogether. There was no appreciable difference between pregnant and nonpregnant sheep in this variability.

Heart Rate and Stroke Volume

The heart rate increased in both pregnant and nonpregnant groups of sheep during the period of the experiment (Fig. 18 and Table 12). However, the increase in the heart rate of the pregnant sheep was greater than that in the nonpregnant sheep (A: $P < 0.01$ and B: $P < 0.02$), and the mean value just before delivery was 109 beats per minute, 30% greater than the mean postpartum value of 84 beats per minute.

The stroke volume at the end of pregnancy was 97 ml, and

Fig. 17 Cardiac output per unit of body weight of the pregnant sheep (upper graph) and the nonpregnant control sheep (lower graph) during approximately the same chronological period.


.20
.18
.16
.14
.12
.10
.08
CARDIAC INDEX L./KG/MIN
BEFORE
PARTURITION
AFTER
90
60
30
0
30
60
DAYS

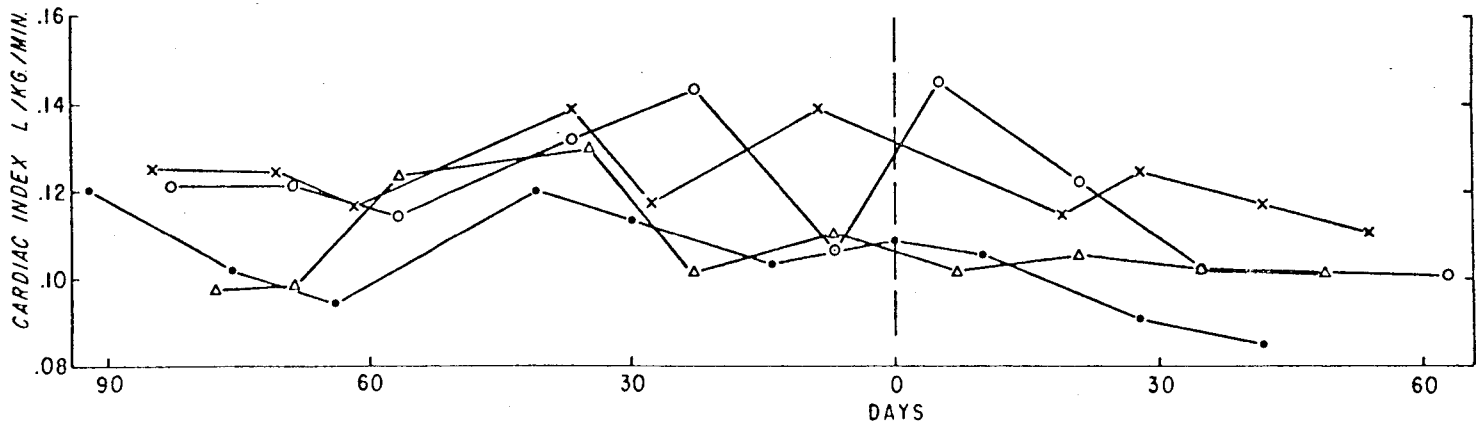
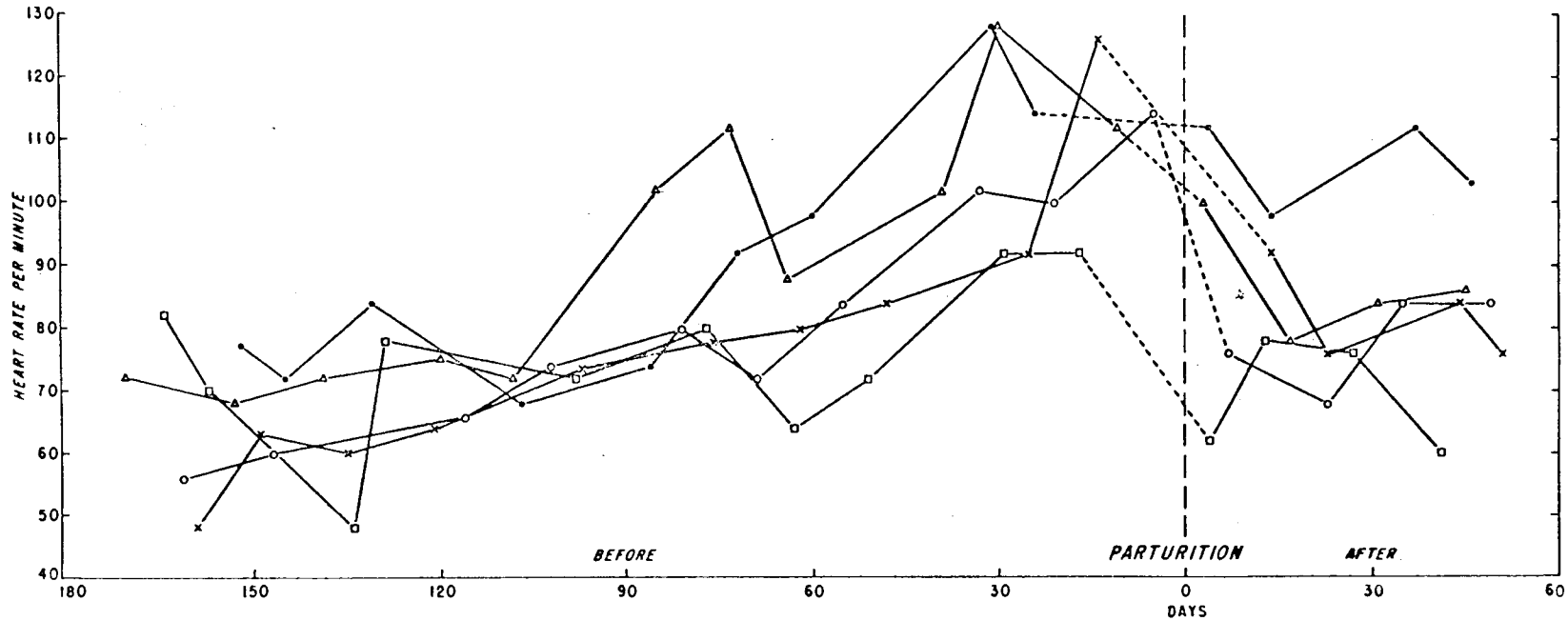

.16
.14
.12
.10
.08
CARDIAC INDEX L./KG./MIN.
90
60
30
0
30
60
DAYS

Fig. 18 Heart rate of the pregnant sheep (upper graph) and the nonpregnant control sheep (lower graph) during approximately the same chronological period.


HEART RATE PER MINUTE
130
120
110
100
90
80
70
60
50
40
BEFORE
PARTURITION
AFTER
180
150
120
90
60
30
0
30
60
DAYS

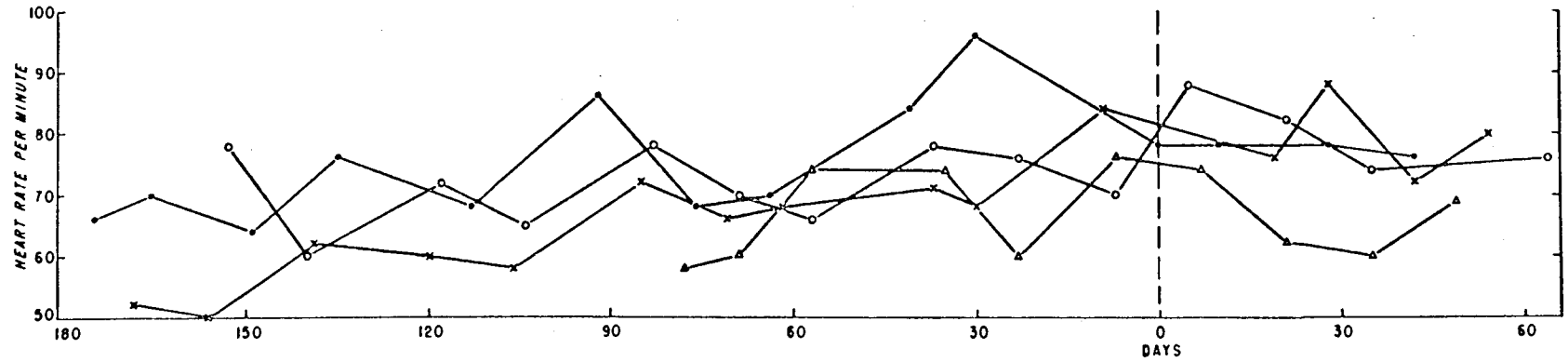

HEART RATE PER MINUTE
100
90
80
70
60
50
180
150
120
90
60
30
0
30
60
DAYS

TABLE 12 AVERAGE HEART RATE AND STROKE VOLUME OF PREGNANT AND NONPREGNANT EWES

Time (weeks)	Pulse rate (beats/min)		Stroke volume (ml)	
	preg.*	nonpreg.+	preg.*	nonpreg.+
Before pregnancy				
3 - 0	67	65	-	-
During pregnancy				
0 - 3	64	66	-	-
4 - 6	70	66	-	-
7 - 9	73	74	-	-
10 - 12	87	67	94	111
13 - 15	84	70	99	108
16 - 18	104	76	90	110
19 - 21	109	77	97	105
Postpartum				
0 - 9	84	75	88	102
Increase above post-partum at term:	30%	3%	10%	3%
Significance level (P):				
Analysis A	< 0.01		N.S.	
Analysis B	< 0.02		N.S.	

*Five pregnant ewes.
+Values for four nonpregnant ewes during the period when the other five were pregnant.

this was 10% greater than the mean postpartum value of 88 ml (Table 12). However, the difference between pregnant and nonpregnant groups was not statistically significant.

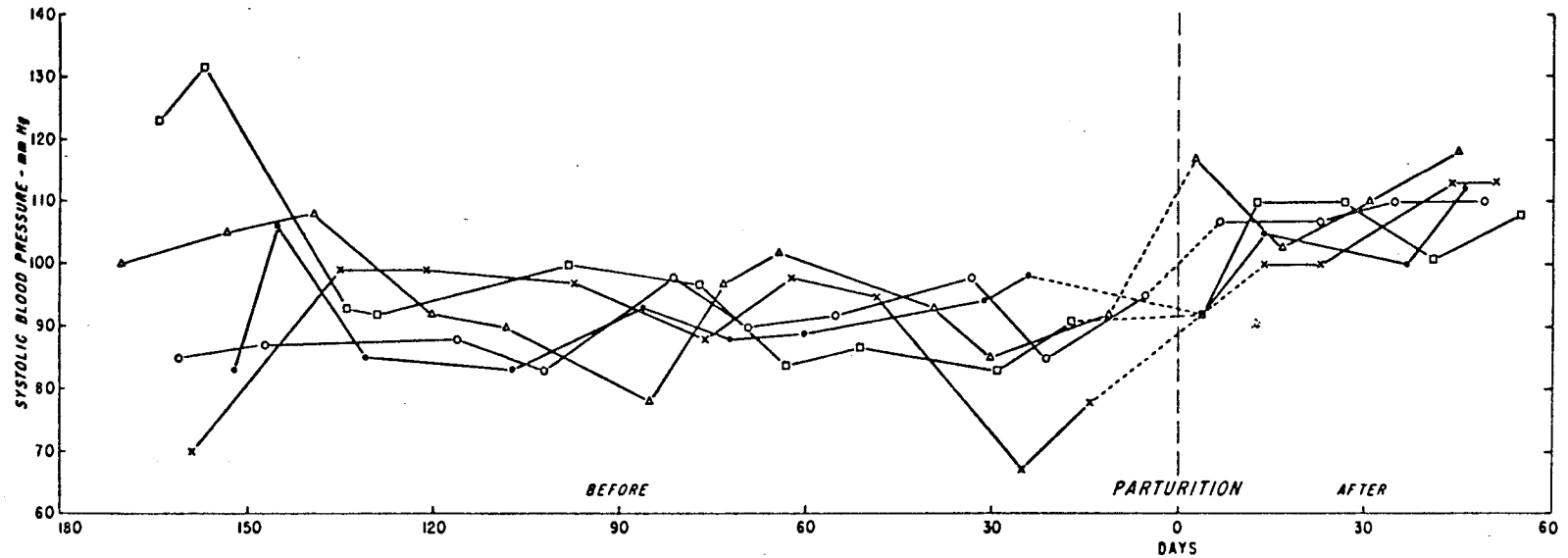
Arterial Blood Pressure

The arterial systolic blood pressure (Fig. 19 and Table 13) increased throughout the period of measurement in the nonpregnant sheep, and decreased somewhat during gestation in the pregnant sheep. This indicates an effective decrease in systolic blood pressure during pregnancy. There was a considerable increase after delivery in the pregnant sheep, the mean value at term being 18% lower than the postpartum value. However, the difference between pregnant and nonpregnant groups only approached significance in Analysis B, when the 30-day postpartum values were subtracted from the 30-day prepartum values. The arterial diastolic blood pressure (Fig. 20 and Table 13) was 15% lower than the postpartum values at term but, as with the systolic pressure, this failed to reach statistical significance.

Peripheral Resistance

The peripheral resistance, calculated from mean arterial blood pressure and cardiac output (Warren and Gorlin, 75, p. 98-99) is shown in Table 14. At term it was 42% lower than the mean

Fig. 19 Arterial systolic blood pressure of the pregnant sheep (upper graph) and the nonpregnant control sheep (lower graph) during approximately the same chronological period.


SYSTOLIC BLOOD PRESSURE - mm Hg
140
130
120
110
100
90
80
70
60
BEFORE
PARTURITION
AFTER
180
150
120
90
60
30
0
30
60
DAYS

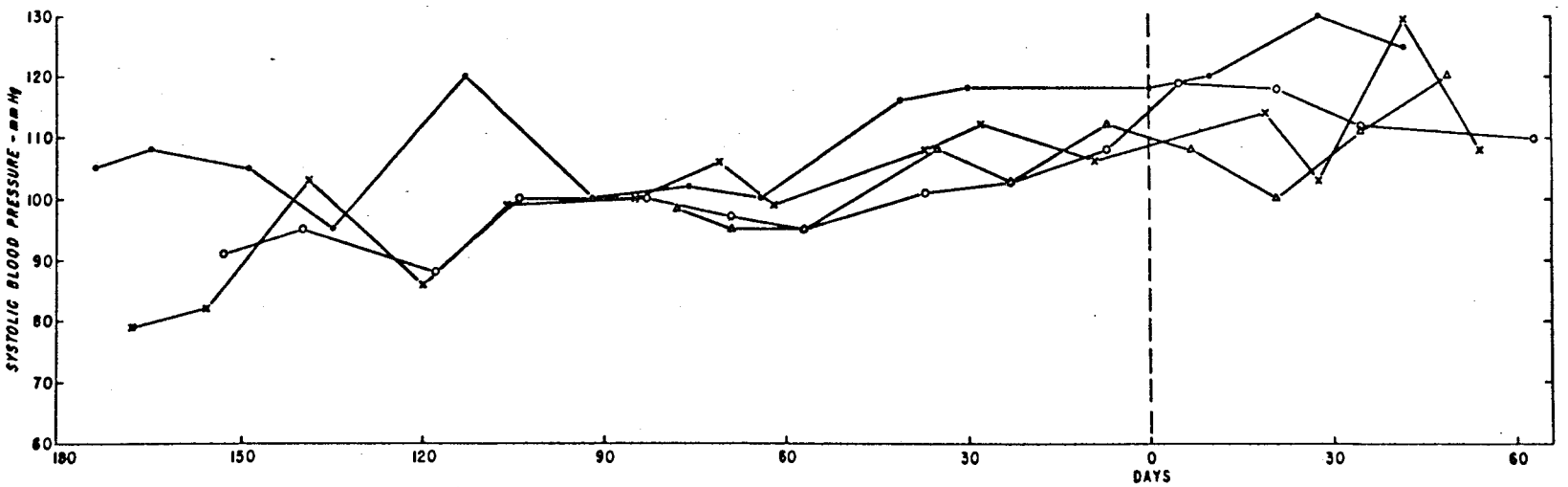
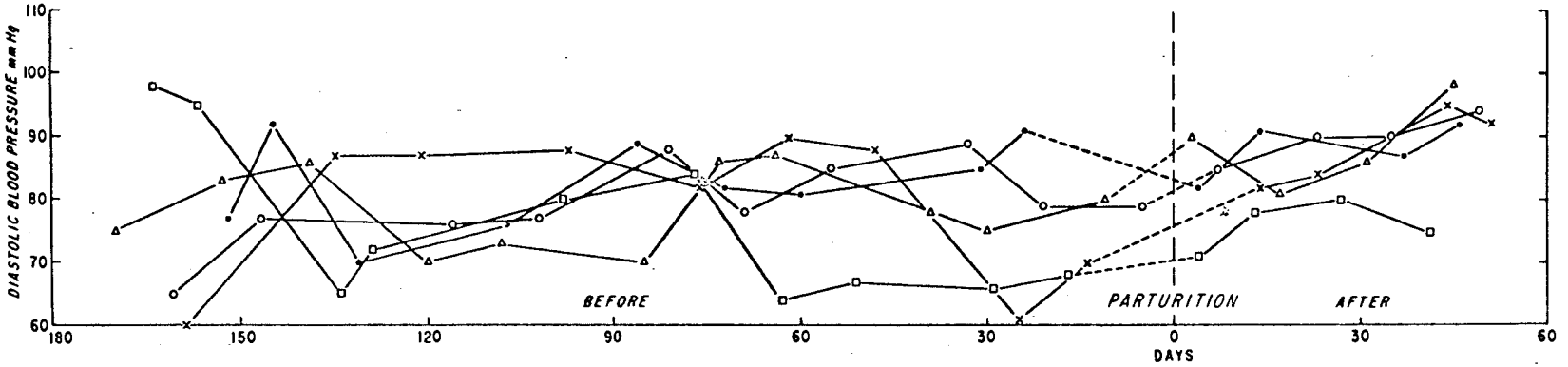

SYSTOLIC BLOOD PRESSURE - mm Hg
130
120
110
100
90
80
70
60
180
150
120
90
60
30
0
30
60
DAYS

TABLE 13 AVERAGE ARTERIAL BLOOD PRESSURE OF PREGNANT AND NONPREGNANT EWES

Time (weeks)	Arterial blood pressure			
	Systolic (mm Hg)		Diastolic (mm Hg)	
	preg.*	nonpreg.+	preg.*	nonpreg.+
Before pregnancy				
3 - 0	94	93	76	78
During pregnancy				
0 - 3	97	98	79	83
4 - 6	91	100	76	87
7 - 9	93	100	81	85
10 - 12	91	101	82	90
13 - 15	93	97	82	84
16 - 18	87	109	76	92
19 - 21	88	111	74	89
Postpartum				
0 - 9	107	116	87	94
Decrease below postpartum at term:	18%	4%	15%	5%
Significance level (P):				
Analysis A	N. S		N. S.	
Analysis B	$\langle 0.1 \rangle$ 0.05		N. S.	

*Five pregnant ewes.
+Values for four nonpregnant ewes during the period when the other five were pregnant.

Fig. 20 Arterial diastolic blood pressure of the pregnant sheep (upper graph) and the nonpregnant control sheep (lower graph) during approximately the same chronological period.


DIASTOLIC BLOOD PRESSURE mm Hg
110
100
90
80
70
60
180
150
120
90
60
30
0
30
60
DAYS
BEFORE
PARTURITION
AFTER

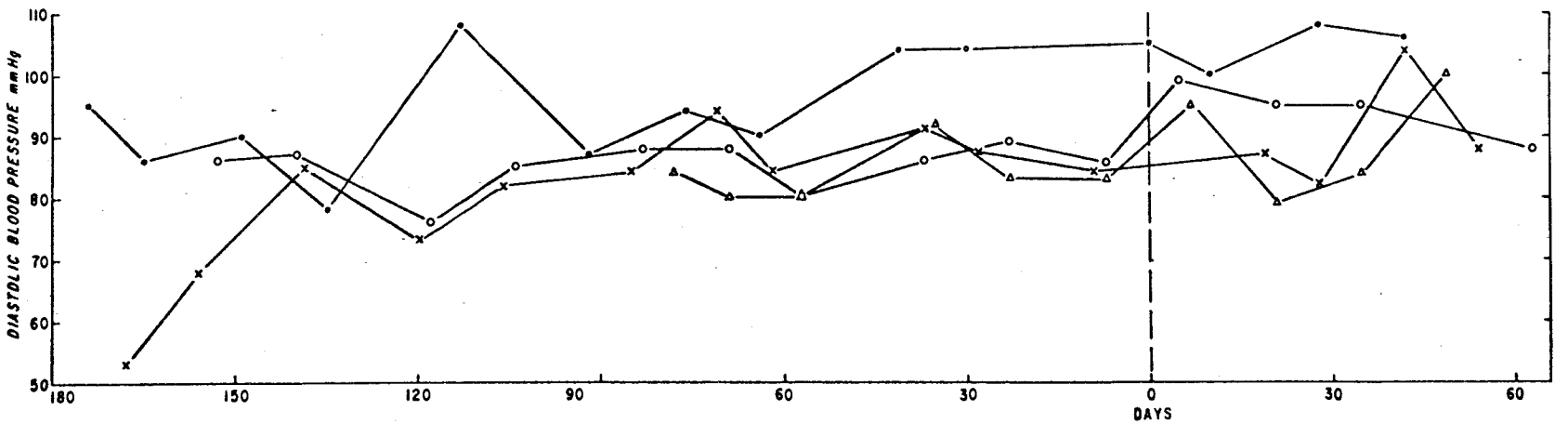

DIASTOLIC BLOOD PRESSURE mm Hg
110
100
90
80
70
60
50
180
150
120
90
60
30
0
30
60
DAYS

TABLE 14 AVERAGE PERIPHERAL RESISTANCE OF PREGNANT AND NONPREGNANT EWES

Time (weeks)	Peripheral resistance dyne sec cm^{-5}	
	Pregnant*	Nonpregnant+
During pregnancy		
10 - 12	868	1024
13 - 15	860	972
16 - 18	724	971
19 - 21	616	958
Postpartum		
0 - 9	1053	1104
Decrease below postpartum at term:	42%	13%
Significance level (P): Analysis A	< 0.02	
Significance level (P): Analysis B	< 0.01	

*Five pregnant ewes.
+Values for four nonpregnant ewes during the period when the other five were pregnant.

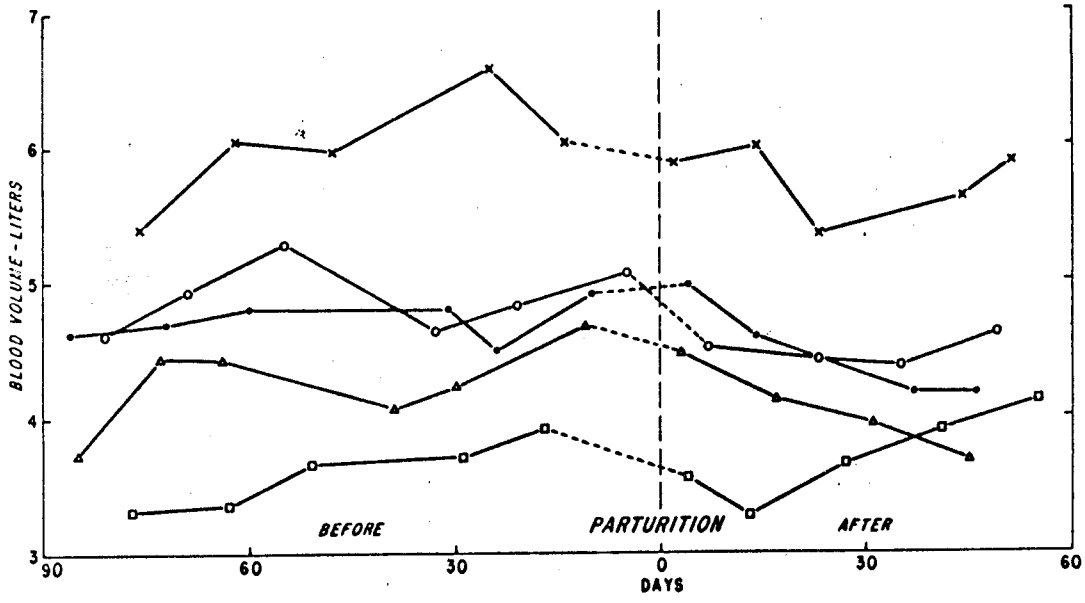
postpartum value. In the nonpregnant sheep the peripheral resistance decreased 13% during the same period, but the difference between the two groups was significant according to both statistical analyses (A: $P < 0.02$ and B: $P < 0.01$).

Blood Volume

The blood volume (Fig. 21 and Table 15) increased by 9% during pregnancy. This was a significant increase according to Analysis B ($P < 0.05$), but was below the 95% level of confidence according to Analysis A. The blood volume per unit of body weight showed no change which could be attributed to pregnancy (Fig. 22 and Table 15). However, it should be remembered that the body weight included both wool growth (the effect of which would cancel out when comparing the two groups) and fetal mass, i.e. fetus, membranes and fetal fluids. This latter weight increase does not add to the mass of the mother from the vascular point of view. Evans blue dye does not cross the placenta, so fetal blood is not measured by it. Thus, if we consider the blood volume per unit of body weight minus the fetal mass, i.e. the maternal "vascularized" body weight with respect to Evans blue, the value probably increases during pregnancy, and there is a hypervolemia.

The mean percent deviation of the single measurements from the mean of the triplicate measurements for a sheep on each

Fig. 21 Blood volume of the pregnant sheep (upper graph) and the nonpregnant control sheep (lower graph) during approximately the same chronological period.


BLOOD VOLUME - LITERS
7
6
5
4
3
BEFORE
PARTURITION
AFTER
90
60
30
0
30
60
DAYS

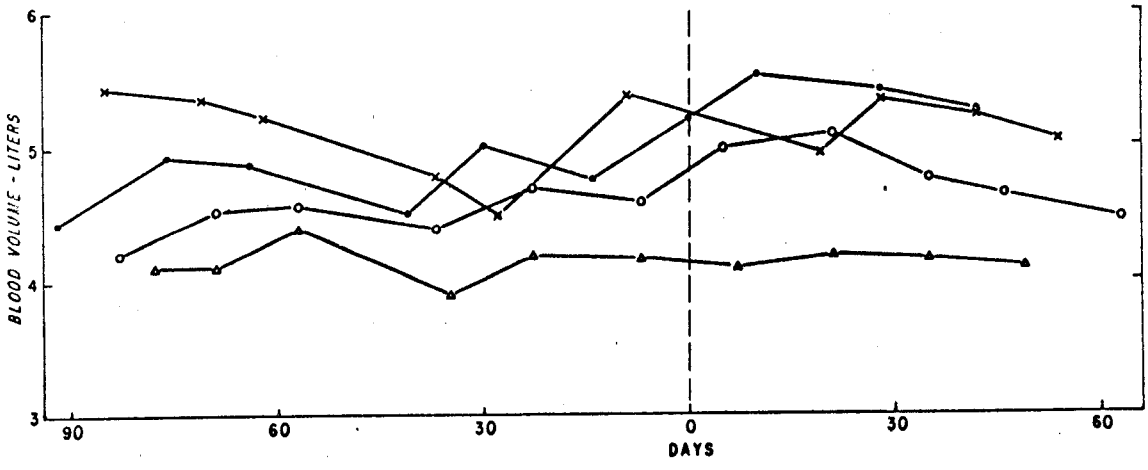
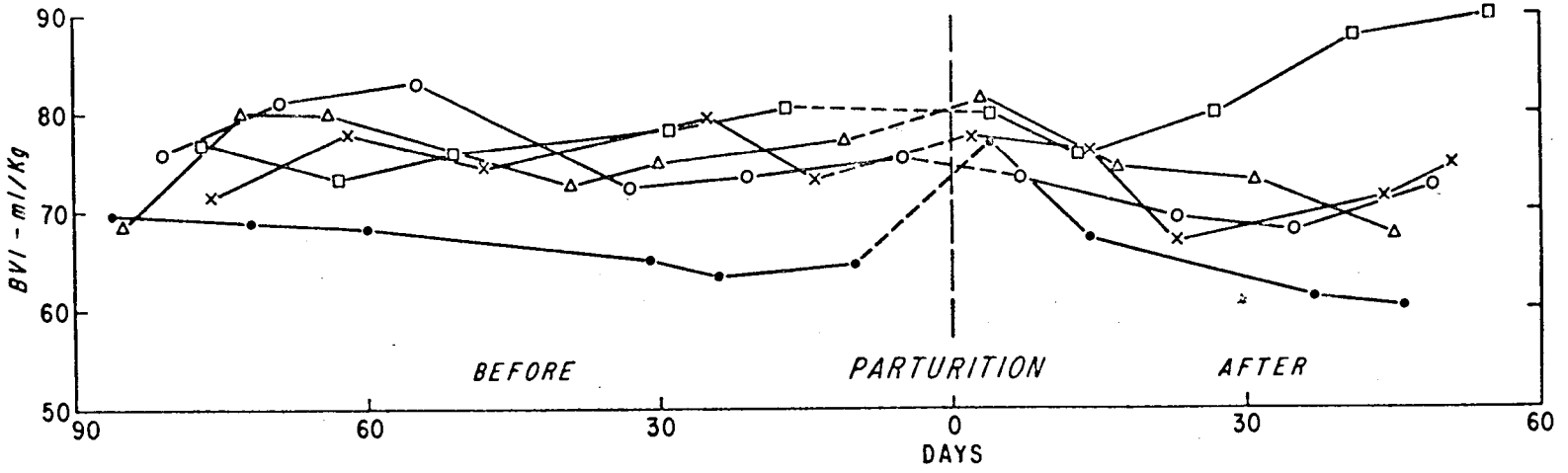

BLOOD VOLUME - LITERS
6
5
4
3
90
60
30
0
30
60
DAYS

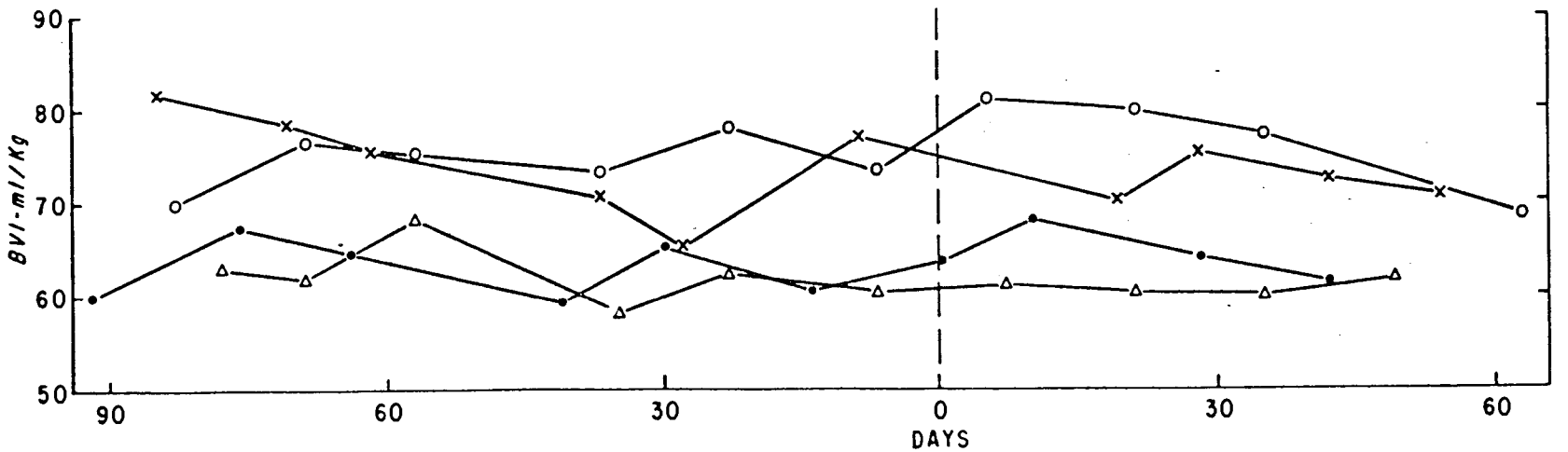
TABLE 15 AVERAGE BLOOD VOLUME AND PLASMA VOLUME OF PREGNANT AND NONPREGNANT EWES

Time (weeks)	Blood volume (liters)		Blood volume (ml/kg)		Plasma volume (liters)	
	preg.*	nonpreg.+	preg.*	nonpreg.+	preg.*	nonpreg.+
During pregnancy						
10 - 12	4.5	4.7	74	70	3.0	3.1
13 - 15	4.8	4.8	77	71	3.1	3.1
16 - 18	4.8	4.5	74	67	3.5	3.2
19 - 21	4.9	4.8	74	68	3.4	3.2
Post-partum						
0 - 9	4.5	4.8	73	68	3.1	3.3
Increase above postpartum at term:	9%	0%	1%	0%	10%	-3%
Significance level (P):						
Analysis A	$\langle 0.1 \rangle 0.05$		N.S.		$\langle 0.01$	
Analysis B	$\langle 0.05$		N.S.		$\langle 0.05$	

*Five pregnant ewes.
+Values for four nonpregnant ewes during the period when the other five were pregnant.

Fig. 22 Blood volume per unit of body weight of the pregnant sheep (upper graph) and the nonpregnant control sheep (lower graph) during approximately the same chronological period.


90
80
70
60
50
BVI - ml/Kg
BEFORE
PARTURITION
AFTER
90
60
30
0
30
60
DAYS


90
80
70
60
50
BVI-ml/Kg
90
60
30
0
30
60
DAYS

experimental day averaged 4.0%. This includes analysis of 90 sets of triplicate determinations of blood volume, or 270 separate determinations in all.

Plasma Volume and Hematocrit

The plasma volume (Fig. 23 and Table 15) increased by about 10% during pregnancy, and this was a statistically significant increase (A: $P < 0.01$ and B: $P < 0.05$).

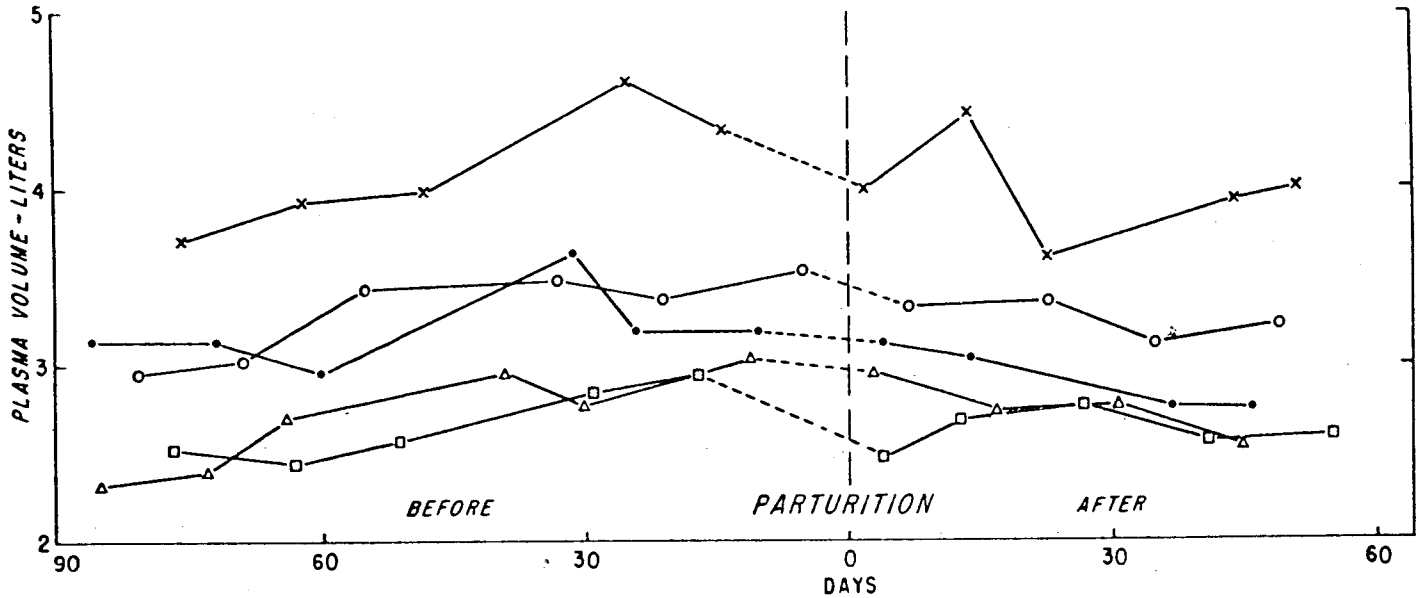
The hematocrit did not alter in any fashion which could be attributed to pregnancy (Fig. 24 and Table 16), and the difference between the groups was not statistically significant. The mean hematocrit in the pregnant group during the experimental period was 32%, and that in the nonpregnant group 33%.

The mean percent deviation of the single measurements from the mean of the triplicate measurements of hematocrit on each experimental day averaged 4.0%. This was calculated from the data of two sheep, representing 18 sets of triplicate determinations.

Variability of the Cardiovascular Changes

The variability of the changes occurring in the pregnant animals is of interest. In Table 17, the percent changes near term of a number of functions of the individual pregnant sheep are shown. Also shown are maternal body weight (mean postpartum) and the birth weight of lambs. There was no apparent relationship between birth weight of the lamb and any of the functions shown. Also there

Fig. 23 Plasma volume of the pregnant sheep (upper graph) and the nonpregnant sheep (lower graph) during approximately the same chronological period.


PLASMA VOLUME - LITERS
5
4
3
2
BEFORE
PARTURITION
AFTER
90
60
30
0
30
60
DAYS

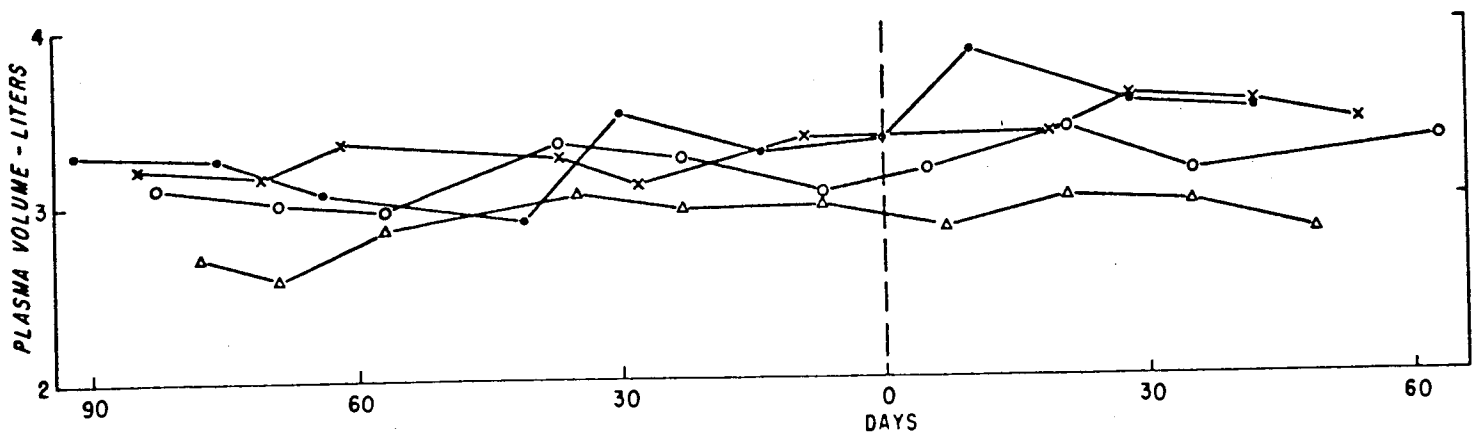
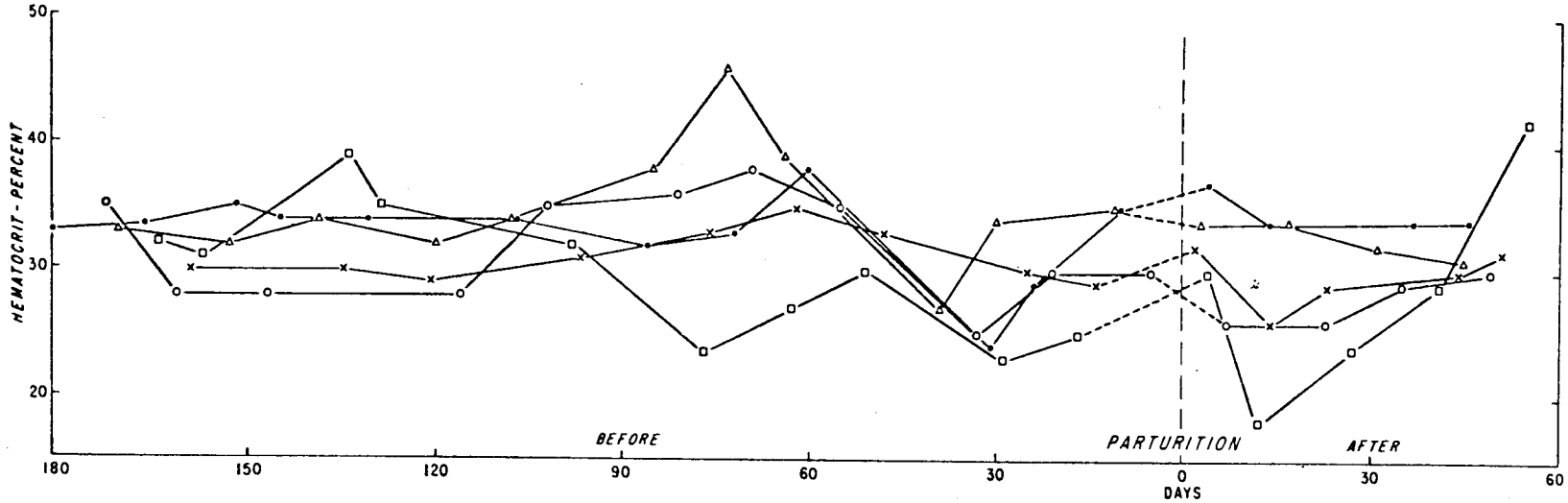

PLASMA VOLUME - LITERS
4
3
2
90
60
30
0
30
60
DAYS

Fig. 24 Hematocrit of the pregnant sheep (upper graph) and the nonpregnant control sheep (lower graph) during approximately the same chronological period.


HEMATOCRIT - PERCENT
50
40
30
20
BEFORE
PARTURITION
AFTER
180
150
120
90
60
30
0
30
60
DAYS

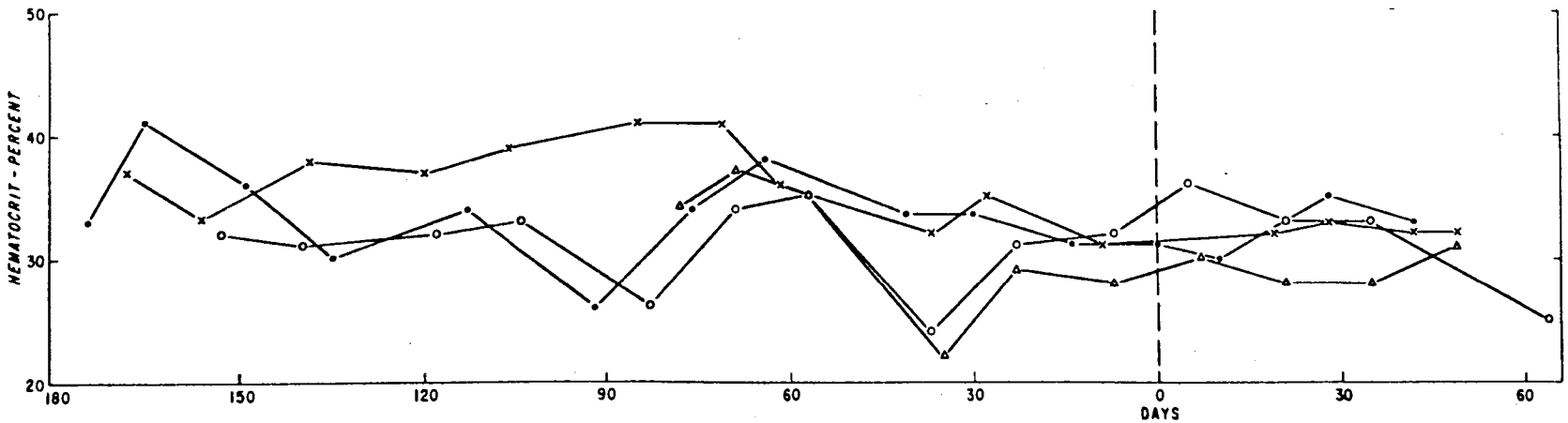

HEMATOCRIT - PERCENT
50
40
30
20
180
150
120
90
60
30
0
30
60
DAYS

TABLE 16 AVERAGE HEMATOCRIT OF PREGNANT AND NON-PREGNANT EWES

Time (weeks)	Hematocrit (%)	
	Pregnant*	Nonpregnant+
Before pregnancy		
3 - 0	31	35
During pregnancy		
0 - 3	33	33
4 - 6	32	35
7 - 9	33	33
10 - 12	34	35
13 - 15	35	36
16 - 18	27	31
19 - 21	31	31
Postpartum		
0 - 9	31	31
Increase above postpartum at term:	0%	0%
Significance level (P): Analysis A	N.S.	
Significance level (P): Analysis B	N.S.	

*Five pregnant ewes.
+Values for four nonpregnant ewes during the period when the other five were pregnant.

TABLE 17 EWE WEIGHT (POSTPARTUM), LAMB WEIGHT AND PERCENT CHANGES IN THE VARIOUS FUNCTIONS DURING PREGNANCY

Sheep No.	Weight (kg)		Percent Changes (30-day postpartum compared to 30-day prepartum values)				
	Ewe	Lamb	Body wt.	Cardiac output	Blood volume	Plasma volume	Peripheral resistance
H300	77	- *	8	37	11	13	-47
I 46	67	3.2*	10	19	-2	3	-6
G137	63	3.2	6	48	11	3	-41
G 11	55	4.5	6	24	5	0	-29
H 62	45	4.5	7	36	9	11	-33

* Lambs born dead

was no apparent relationship between body weight of the ewe and the percent change of any of the measured cardiovascular functions. As might be expected, the change in cardiac output is related to the change in peripheral resistance. Cardiac output is a component of peripheral resistance, so this is not unexpected. Change of cardiac output was positively related to the change of blood volume ($\underline{r} = 0.2$) but this was not statistically significant.

V DISCUSSION

Cardiac Output

The changes occurring in the cardiovascular system of ewes during pregnancy which are reported here have many similarities to those occurring in the human. Cardiac output increases 40-50% in both species, and a similar increase was reported in the pregnant bitch (Table 1). In the human, however, the maximum value occurs at about the seventh lunar month, and cardiac output then declines. This decrease was not evident in the sheep, but because the period of gestation in the sheep (21 weeks) is only half that in the human, it is possible that more frequent observations would need to be made to recognize this reversal if it actually does occur.

An examination of other workers' values for cardiac output gives an indication of the general accuracy of the method used in this work. The mean value of the cardiac output for the nonpregnant sheep in this work was 115 ml/kg/min. Schambye (63, P. 1159-1160) reported an average cardiac output of 112 ml/kg/min (range 80-157) in 11 unanesthetized standing sheep (n = 37) using the dye dilution method. Stowe and Good (69) recorded 126 ml/kg/min (range 94-164) in six ewes anesthetized with pentobarbital, and later 104 ml/kg/min (range 72-137) in five anesthetized sheep (70)

using the direct Fick technique. Hamlin and Smith (32), using eight small sheep (mean weight 28 kg) anesthetized with pentobarbital, measured a mean cardiac output of 131 ml/kg/min (range 103-204) with the dye dilution technique. The extent to which anesthesia and a recumbent position alters the cardiac output is rather uncertain, though Stowe and Good (69) state that the differences observed in cardiac output measurements between anesthetized and unanesthe- calves were insignificant. It seems probable that the values obtained on trained standing sheep, as in Schambye's report and the present work, reflect values more closely related to normal physiological function than those obtained on anesthetized sheep. Support was given to the concept of a "basal" or "standard" physiological state by the reproducibility of triplicate measurements. The mean deviation from the mean during triplicate measurements of cardiac output was 5.9% in the present work. This indicates the remarkably good agreement between three measurements made at ten-minute intervals. It does not, however, give a measure of the spread of values obtained. The accuracy of the dye dilution technique is generally considered to be about 10 or 20% (5; 65). The mean percent deviation from the mean for blood volume and hematocrit was 4.0%. These may be expected to be more stable functions over ten-minute periods than the cardiac output, and so this 4.0% is

probably mainly a measure of technical error, e. g. in injection of dye, blood sampling, spectrophotometry, etc.

The increase of cardiac output during pregnancy above postpartum control levels varied in different individuals from 19-48%. Attempts to correlate the percent change in cardiac output with the body weight of the fetus, body weight of the ewe, or any other measured vascular function were in vain. This does not preclude such a relationship because the number of animals used was small, and they were not uniform in breed or age. The variability in recorded cardiac output increase may, therefore, be attributed to environmental factors such as climate, nutrition and unrecognized ailments, and variability due to temperament, rather than to changes solely related to reproductive function.

The distribution of the increased cardiac output is of interest. It can be partly accounted for by extra blood flow to the pregnant uterus. Near term in anesthetized pregnant sheep and goats weighing about 40 kg this amounts to approximately 1. 5 liters per minute (Metcalfe, Romney, Swartwout, Pitcairn, Lethin and Barron, 51; Huckabee, Metcalfe, Prystowsky and Barron, 39). Similar values for uterine blood flow have been reported in unanesthetized, standing sheep (Metcalfe, Huckabee, Prystowsky, Hellegers, Meschia, Wolkoff and Barron, 50). Thus one could

assume a uterine blood flow of about two liters per minute in the sheep (approximately 60 kg) in the present report. There remains an increment in cardiac output of approximately one liter per minute to be accounted for in these sheep.

Burt (11) demonstrated an increase in forearm blood flow in women in the latter part of pregnancy. Blood flow through the hands was also shown to increase (Abramson, Flachs and Fierst, 1). Herbert, Banner and Wakim (35) demonstrated an increase in blood flow in the extremities in the latter half of human pregnancy, with a decline after 38 weeks. These increases in blood flow to the extremities have been related to the need for greater heat loss, because of the rapidly metabolizing fetus. This has not yet been critically examined in the human. If the pregnant ewe has a similar requirement, one might expect an increase in blood flow to the upper respiratory tract because evaporation from the respiratory passages is an important avenue of heat loss in the heat stressed sheep (Bligh, 6). The respiration rate was 22% higher at term than postpartum in these sheep (unpublished data), but this was not statistically significant. However, the values obtained were variable, and it may be possible to recognize an increase in respiration rate under more stable conditions, or in a warmer environment. This requires further study.

Linzell (46, p. 500) measured blood flow through the mammary glands of goats. The weight of a single mammary gland of a nonlactating goat increased greatly in the last 2-1/2 months of pregnancy from about 0.25 kg to 1.5 kg. Blood flow increased more than fivefold during this period to a maximum of 0.5 L/min at delivery. The goat presumably had a much higher milk yield and udder size than the sheep used in the present experiment. Despite this, it seems that the udder of sheep would be an important recipient of the increased cardiac output near the end of gestation. This suggestion is supported by the report of Cloete (17, p. 530-532) that mammary gland weight increases from about 200 gm early in pregnancy to 800 gm in the fifth month of pregnancy in the Merino ewe. Furthermore, he reported that towards the end of the second month of gestation the diameter of the mammary blood vessels (external pudic arteries and veins and subcutaneous abdominal veins) was doubled.

The changes occurring in renal blood flow in the human during pregnancy are still unclear. Sims and Krantz (64) noted an increase in renal plasma flow in women of about 200 cc/min. This declined after 32 weeks to normal nonpregnant control values. The blood flow through the kidneys probably increased about 300 cc/min in these pregnant women. DeAlvarez et al. (19) reported a very

early increase in renal blood flow of about 700 cc/min, and then a persistent decline to values below control levels after 28 weeks. Both the above reports show an increase in renal blood flow in pregnant women, but the exact behavior of this function during pregnancy is still a matter for further study. Regarding other organs, Munnell and Taylor (54) were unable to disclose any change in liver blood flow during pregnancy; nor was any change noted in cerebral blood flow during pregnancy (McCall,47). Further support of the idea that there are alterations in blood vessels in pregnancy is the observation (Burwell and Metcalfe, 13, p. 18-19) that spider angiomata commonly occur in pregnant women and that arterial aneurysms may rupture during pregnancy.

The above discussion suggests the probable distribution of the increased cardiac output in pregnant sheep, i.e. (a) uterus, (b) periphery and/or upper respiratory tract, (c) mammary glands, (d) kidneys. For the latter three areas, however, information is lacking in the sheep.

The comparative aspects of cardiovascular changes during pregnancy are of great interest, particularly with respect to the type of placenta. It has been pointed out (Bartels, Moll and Metcalfe, 4, p. 1729) that oxygen supply to the fetus can be increased by an increase in maternal placental blood flow. It is thus possible that

increased maternal placental blood flow may be a species adjustment to various placental types. Those with the five-layered syndesmochorial placenta (e. g. sheep) may require a greater placental blood flow to attain equivalent fetal oxygenation compared with those with the three-layered hemochorial placenta (e. g. man), because of the greater diffusion distance. Such an increase in placental blood flow would be reflected in increased cardiac output. However, critically obtained information is inadequate and these theories remain conjectural.

Heart Rate and Stroke Volume

The increased cardiac output was brought about in humans by an increase in heart rate and stroke volume (see Tables 2 and 3). This also appeared to be the case in the sheep used in this experiment, though the increase in heart rate was predominant.

Blood Pressure and Peripheral Resistance

The striking decrease in peripheral resistance which was described for the human (see Table 5) also occurred in the sheep. The heart output increased during pregnancy with no increase in arterial blood pressure, in fact with a possible decrease in blood pressure. Some possible explanations for this remarkable physiological fact are described below.

Blood Volume, Plasma Volume and Hematocrit

When comparing the present results in sheep with those in humans the most divergent results occur in the blood volume and its constituents (see Tables 6, 7 and 8). In humans there was a 27% increase in blood volume, while in sheep it was found to be only 9%. Furthermore, the human data consistently show a decrease in hematocrit during pregnancy, while this was absent in the sheep in the present experiment. Barcroft et al.'s experiment (3), however, illustrated an increase in blood volume of sheep during pregnancy comparable to that in the human (24%) and also a considerable decrease in hematocrit (24%). The discrepancy between the two results is unexplained. One may postulate that the absence of a decrease in hematocrit in the present experiment could be attributed to the regular administration of iron. Hytten and Duncan (40, p. 861) in challenging the view that "anemia of pregnancy" is due to an iron deficiency, state that blood formation can be stimulated in healthy nonpregnant persons by giving iron in large doses so the response to iron in pregnant women is not sound evidence that they are suffering from iron deficiency. They state that no advantage has been demonstrated in increasing the amount of red cells in pregnancy, and in fact it may even be undesirable because of increased blood viscosity and resistance. Whether this theoretical argument is equally valid

for the sheep is open to discussion, as the nonpregnant sheep normally has a lower hematocrit than the human.

The present experiment does not seem to lend support to the theory that hypervolemia causes the cardiodynamic changes in pregnancy. This theory rests on the idea that hypervolemia increases venous return, which in turn results in increased cardiac output. However, in the present experiment the blood volume increased only 9%, while the cardiac output increased by 40%, thus suggesting no causal relationship.

It is possible that species differences occur regarding anemia of pregnancy. It may be of significance that in the human, rabbit and rat, this condition was reported by all workers, while in the ungulates three of four experiments demonstrated no change in hematocrit during pregnancy. Whether this apparent difference is concerned with placental type or hormone balance, particularly estrogen and progesterone as suggested by Zarrow (80, p. 992), is not known. Indeed, the species differences require more confirmation before they can be considered to be established.

Possible Mechanisms Involved in Cardiovascular Changes in Pregnancy

The current theories regarding the mechanisms involved in bringing about the cardiovascular changes during pregnancy were discussed in II Review of the Literature. This section will attempt to

summarize, and to consider which of these theories are supported by the present experiment.

The decrease in peripheral resistance in these sheep suggests that the uterus is an area of low vascular resistance in pregnancy. This is supported by the high values for uterine blood flow (Metcalfe et al., 50; 51) in this species. While the uterus appears to be the major area of decreased resistance in the pregnant female, the extent to which resistance in other organs decreases is not known. However it seems likely that blood flow to the mammary gland increases, as a high rate of flow has been reported for the goat (46, p. 500). Probably blood flow through the kidneys, and the periphery, and the upper respiratory tract increases, if we can extrapolate from the human. If the pregnant uterus is in fact an area of low vascular resistance, how is this brought about? The prime stimulus for the increased growth and vascularity of the uterus seems to be estrogen, though this has yet to be demonstrated conclusively in sheep. The increased vascularity alone would presumably result in an increase in blood volume, because of the increase in vascular space. A further increase could be brought about by a substance with salt and water retaining ability, such as estrogen. However, in the latter case there would need to be some concomitant influence on vascular tone or increase in vascular capacity to

prevent an increase in blood pressure. Possibly estrogen is also responsible for this (Parer, Metcalfe and Jones, 57). Experimental evidence (Parer, Metcalfe and Moll, unpublished data) suggests that progesterone alone is not responsible, though an interaction of estrogen and progesterone needs to be examined.

The experiment indicates the complexity of the adjustments which occur in the ewe in pregnancy. Presumably these changes in cardiac output and other functions are necessary for the successful production of healthy offspring. An important question suggested by this study is the extent to which the cardiovascular adjustments of pregnancy can vary without affecting fetal development. This question remains to be answered.

VI. SUMMARY

The cardiovascular adjustments accompanying pregnancy were studied in a group of five pregnant ewes. Similar measurements were carried out on a group of four nonpregnant ewes to evaluate environmental changes. Measurements were made on the animals without narcosis at approximately two to three week intervals, and continued for nine weeks following delivery.

The cardiac output increased during pregnancy reaching an average value of 10.3 liters/minute in the last three weeks. This was 41% greater than the average postpartum value, and the difference was statistically significant ($P < 0.01$). The average values of the other measurements in the last three weeks of pregnancy were as follows:

a. Cardiac output per unit of body weight, 157 ml/kg/min. This was 31% greater than the postpartum average ($P < 0.02$).

b. Heart rate, 109 beats per minute. This was 30% greater than the postpartum average ($P < 0.02$).

c. Arterial systolic blood pressure, 88 mm Hg. This was 18% less than the postpartum average ($P < 0.1$).

d. Arterial diastolic blood pressure, 74 mm Hg. This was 15% less than the postpartum average (N.S.)

e. Peripheral resistance, 616 dyne sec cm^{-5}. This was 42%

less than the postpartum average ($P < 0.01$).

f. Blood volume, 4.9 liters. This was 9% greater than the postpartum average ($P < 0.05$).

g. Blood volume per unit of body weight, 74 ml/kg. This was 1% greater than the postpartum average (N. S.).

h. Plasma volume, 3.4 liters. This was 10% greater than the postpartum average ($P < 0.05$).

i. Hematocrit, 31%. This was the same as the postpartum average (N. S.).

j. Body weight, 66.8 kg. This was 8% greater than the postpartum average ($P < 0.01$).

The average maximum value of cardiac output in the last three weeks of pregnancy varied from 19-48% above postpartum values in individual sheep. There was no correlation between this function and birth weight of lambs or body weight of the ewes; nor was there any evident relationship between these latter values and changes in any of the other cardiovascular functions.

These changes in the cardiovascular system of ewes were similar in many ways to those previously reported for the human. Exceptions were the smaller increase in blood volume and absence of "anemia of pregnancy" in the sheep. The possible distribution of

the increased cardiac output and the mechanisms bringing about the cardiovascular adjustments to pregnancy are discussed.

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