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THE PATHOLOGICAL PHYSIOLOGY OF HEARTWATER [*COWDRIA (RICKETTSIA) RUMINANTIUM*—COWDRY, 1926]

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INTRODUCTION

Henning (1949) summarized the publications on heartwater. A study of the literature cited by him and subsequent publications indicated that little attention had been paid to the pathological physiology of the disease. The only report of work on the chemical pathology of the disease which could be found was that by Graf (1933) who confined himself to the determination of a limited number of blood constituents without finding any significant changes. It was, therefore, decided to follow the pathological physiology of the disease in as great detail as possible in the hope that the findings might indicate rational lines of supportive treatment. This aspect is of practical importance as many animals fail to recover in spite of specific chemotherapy which is judged successful on the criterion of body temperature.

METHODS

In all, 15 adult Merino sheep and two Friesland oxen were infected with heartwater by the intravenous route, using infected sheep blood, and the ensuing reactions studied. The methods used for the blood analyses are listed below.

Plasma Na and K....	Clark (1959).
Plasma Mg.....	Neil & Neely (1956).
Plasma Cl.....	Method of Schales and Schales: in Hawk, Oser & Summerson (1954).
Blood Ca.....	Ferro & Ham (1957).
Inorganic P.....	King & Wootton (1956).
Haemoglobin.....	King & Wootton (1956).
Plasma Proteins.....	Method of Kingsley: in Hawk, Oser & Summerson (1954).
Blood Sugar.....	Folin-Wu Principle: in Hawk, Oser & Summerson (1954).
Blood Urea N.....	Brown (1957).

In five of the sheep and both cattle the carotid artery on one side of the neck was exteriorized and placed in a tube of skin for a length of about three inches. This enabled the systolic and diastolic blood pressures to be taken by the conventional auscultatory method using a rubber cuff about 1½ inch in width. The reduced width of the cuff may have given somewhat high readings but this factor was standard throughout.

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RESULTS

A.—Sheep

Reactions

The incubation period varied from seven to 11 days (average nine), the onset of the disease being taken as from the first rise in temperature to above 104 F. The duration of the disease was three to eight days. All the cases produced were fatal. The body temperature remained between 104 and 106 °F until a final drop before death. None of the animals showed a biphasic temperature reaction, neither did they show the well-known nervous symptoms of heartwater. This may have been connected with the fact that the animals were housed throughout the experiment.

Food intake

The food intake was measured on four sheep. There was a rapid fall in food intake from the onset of fever with complete inappetence after the third day.

Water and sodium balance

The same four sheep were kept in metabolism cages and the water intake and urine volume measured. They were dosed 1 gm. NaCl per day.

TABLE 1.—*Water and sodium balance*

	Days of reaction					
	0	1	2	3	4	5
Water intake (l).....	2·8	2·2	2·6	1·9	1·4	0·7
Urine volume (l).....	0·76	0·74	—	0·72	0·43	0·33
Na output (M. eq.).....	27	44	—	40	26	11

(Figures represent average of four sheep).

The fact that, for the first three days, the water intake decreased more rapidly than did the urine output indicates a loss of total body water during the initial stages of the disease, if a constant extrarenal water loss is assumed. This is also indicated by the increased sodium loss recorded on days 1 and 3 despite a reduced food intake. The subsequent fall in urinary sodium output was probably a reflection of faecal loss as it coincided with the appearance of soft pultaceous faeces.

Post mortem findings

The post mortem findings were typical of heartwater except that there were only small amounts of fluid in the body cavities and moderate oedema of the lungs. The diagnoses were confirmed by the demonstration of *C. ruminantium* in impression smears made from the brain.

Haematology

The red cell counts and haemoglobin content fluctuated in accordance with the haematocrit, indicating that there were no marked changes in mean cell volume or haemoglobin content. (The haematocrits are discussed later).

No marked changes were found in the total or differential leucocyte counts with the exception that the eosinophiles disappeared from the circulation even before the first rise in temperature.

Blood chemistry

The large number of figures obtained have been condensed and given in Table 2 as averages.

TABLE 2.—*Blood chemistry*

	Days of reaction				
	Prior	1 & 2	3 & 4	5 & 6	7 & 8
Plasma Na (m. eq./l).....	139 (7)	135 (4)	138 (5)	136 (5)	132 (1)
Plasma K (m. eq./l).....	5·3 (7)	5·4 (6)	5·1 (6)	5·2 (3)	3·5 (1)
Plasma Mg (m. eq./l).....	1·9 (4)	1·3 (3)	1·2 (4)	1·1 (2)	—
Plasma Cl (m. eq./l).....	106 (7)	100 (7)	102 (7)	97 (4)	107 (1)
Plasma HCO_3 (m. eq./l).....	25·8 (3)	28·6 (3)	20·5 (5)	10·28 (3)	—
Blood Ca (mg. %).....	11·4 (7)	12·3 (4)	12·9 (3)	9·4 (3)	—
Inorganic P (mg. %).....	4·6 (7)	3·8 (4)	2·6 (3)	5·1 (4)	—
Blood Urea N (mg. %).....	24 (4)	38 (1)	40 (3)	35 (1)	—
Creatinine (mg. %).....	0·8 (6)	0·5 (3)	0·5 (6)	0·5 (3)	0·5 (1)
Hb (gm. %).....	11·6 (5)	11·2 (4)	12·7 (5)	12·1 (1)	13·5 (1)
Total Plasma Protein (gm. %)	6·6 (13)	6·9 (8)	7·0 (9)	6·6 (5)	5·8 (1)
Albumen (gm. %).....	4·0 (13)	3·8 (8)	4·5 (7)	4·0 (5)	4·0 (1)
Globulins (gm. %).....	2·6 (13)	3·1 (8)	2·5 (7)	2·6 (5)	1·8 (1)

(The figures in brackets represent the number of readings).

None of the blood constituents listed in the table showed any constant changes during the course of the disease. The marked drop in plasma bicarbonate was probably due to the extreme hyperpnoea which accompanied the hyperthermia. As mentioned above, lung oedema was not marked in these cases. Should it occur, a respiratory acidosis could be expected.

The serum copper and magnesium contents were determined in four reacting sheep and found to be within normal limits.

The fact that the blood creatinine showed no rise indicates the absence of serious kidney damage. The slight rise in blood urea may be due to circulatory failure.

No evidence of liver damage was found. There was no bilirubinaemia and the " Bromsulphalein " retention test was negative in three sheep each in an advanced stage of the disease.

Blood sugar

The blood sugar level was followed in ten sheep. The figures remained within normal limits (40–55 mg. per cent) until some 24 hours before death. Five of the ten sheep showed a marked terminal rise in blood sugar, from 60 to 93 mg. per cent being recorded. In the other five sheep the figures from samples taken within 24 hours of death were normal. A terminal rise in blood sugar in sheep is by no means peculiar to heartwater as it has been found in pregnancy disease, tribulosis and enterotoxaemia. In these diseases, as in heartwater, a concurrent terminal glycosuria is often seen.

Plasma colour

It was noted that, after the onset of the reaction, the plasma became progressively darker in colour often reaching a deep orange. The van den Bergh test on these samples consistently showed the presence of less than 1 mg. per cent total bilirubin which was far too little to account for the plasma colour. The nature of the pigment is as yet unknown but its presence probably accounts for the yellow colour of the effusions often seen in heartwater.

Red cell sedimentation rate

This was taken in Wintrobe tubes at an angle of 45° over one hour. The average sedimentation rate before infection was 3·5 mm. (2 to 6) which rose to 7·0 mm. (5 to 8) on the first day of infection and 17·5 (15 to 20) on the fifth day.

TABLE 3.—*Carotid artery systolic and diastolic pressures.*

	Day of reaction				
	Prior	2nd	4th	5th (a.m.)	5th (p.m.)
6.....	120/90	130/90	130/100	126/85	115/0
7.....	—	150/100	130/80	70/40	—
8.....	115/82	140/95	110/80	—	—
9.....	123/90	112/90	110/90	125/0	—
10.....	105/80	126/80	92/70	122/0	—

Sheep numbers correspond with those in Table 4

Arterial blood pressure

In all the above cases the animal died within 24 hours of the last reading being taken. It will be noted that there was a very marked terminal drop in diastolic pressure in the terminal stages. By a diastolic reading of " 0 " is meant that a loud thumping sound was heard over the artery with no pressure being applied to the cuff. During this stage the pulse was bounding and typically " water hammer " in nature. So great was the pulse pressure that the exteriorized " arterial loops " could be seen bounding at each pulsation and the pulse rate could be counted from a distance with ease. At the same time the jugular veins were distended. When bleeding, a high venous pressure was indicated by the fact that the blood spurted from the needle and often forced back the plunger of the syringe without proximal occlusion of the vein. The heart sounds were exceedingly loud but normal in character.

Plasma and blood volumes

The plasma volume was determined by the Evan's blue method. Sheep were given 15 mg. and the cattle 30 mg. of the dye. Samples were taken ten and 20 minutes after injection and zero concentration estimated by backward extrapolation, assuming a linear decline in concentration.

TABLE 4.—*Haematocrit, plasma and blood volume*

Sheep No.		Prior	Days of reaction						
			1	2	3	4	5	6	8
1	Hc.....	36	—	31	37	48	—	—	—
	P.V.....	1·46	—	1·92	—	0·50	—	—	—
	B.V.....	2·52	—	2·91	—	0·96	—	—	—
2	Hc.....	40	34	—	—	31	34	38	43
	P.V.....	1·55	—	—	—	1·03	—	0·87	1·07
	B.V.....	2·58	—	—	—	1·49	—	1·34	1·90
3	Hc.....	47	—	—	35	32	43	—	—
	P.V.....	1·69	—	—	—	1·63	0·24	—	—
	B.V.....	3·15	—	—	—	2·40	0·42	—	—
4	Hc.....	37	35	—	—	42	46	—	—
	P.V.....	1·50	1·82	—	—	1·38	0·74	—	—
	B.V.....	2·37	2·80	—	—	2·38	1·37	—	—
5	Hc.....	36	—	30	27	—	35	—	—
	P.V.....	—	—	2·22	1·40	—	1·09	—	—
	B.V.....	—	—	3·17	1·92	—	1·70	—	—
6	Hc.....	27	22	—	—	11	10	—	—
	P.V.....	2·46	2·50	—	—	2·17	2·02	—	—
	B.V.....	3·28	3·21	—	—	2·40	2·23	—	—
7	Hc.....	35	26	—	—	12	12	—	—
	P.V.....	2·17	2·14	—	—	2·46	1·97	—	—
	B.V.....	3·19	2·89	—	—	2·80	2·24	—	—
8	Hc.....	36	28	—	—	28	—	—	—
	P.V.....	2·08	1·97	—	—	1·97	—	—	—
	B.V.....	3·15	2·74	—	—	2·74	—	—	—
9	Hc.....	35	—	33	—	32	33	—	—
	P.V.....	2·27	—	2·38	—	1·95	2·38	—	—
	B.V.....	3·44	—	3·60	—	2·87	3·55	—	—
10	Hc.....	34	—	30	—	25	27	—	—
	P.V.....	2·05	—	2·30	—	2·00	2·14	—	—
	B.V.....	3·06	—	2·90	—	2·64	2·93	—	—

Hc = Haematocrit per cent.
P.V. = Plasma volume (litres).
B.V. = Blood volume (litres).

In all cases shown in Table 4 the animal died within 24 hours of the last readings shown. It will be seen that, with the exception of sheep No. 6, 8 and 10, there was a marked drop in plasma and blood volume shortly prior to death. That this occurred suddenly and terminally is well shown by the figures for sheep No. 3 and 4 where readings were taken on the day prior to death and shortly before death.

Haematocrits

As will be seen from Table 4 there was a general tendency for the haematocrits to fall, often sharply, during the disease and rise suddenly terminally. This last rise coincided with the drop in plasma volume to which it can be partly attributed.

B.—Cattle

The incubation periods in the cases of the two calves were 15 and 16 days respectively, death taking place in four and two days after the first rise in temperature. Both animals showed the typical nervous symptoms of heartwater.

TABLE 5.—*Clinical findings. Cattle*

Day of reaction	Bov. 1			Bov. 2	
	Prior	3rd	4th	Prior	2nd
Haematocrit (%)	27	15	16	34	29
Red cell sed. rate (mm./hr.)	5	13	10	2	5
Plasma Na (m. eq./l)	138	144	144	132	132
Plasma K (m. eq./l)	5·4	5·2	5·5	4·5	5·5
Plasma bicarb. (m. eq./l)	36	35	28	36	32
Total plasma proteins (gm. %)	7·0	6·0	6·2	8·1	7·0
Albumen (gm. %)	1·4	2·8	2·6	2·8	2·4
Globulins (gm. %)	5·6	3·2	3·6	5·3	4·6
Blood sugar (mg. %)	37	46	72	36	55
Creatinine (mg. %)	0·8	1·1	1·1	1·0	1·2
Total bilirubin (mg. %)	0	0	0	0	0
Plasma volume (litres)	6·25	7·14	6·82	6·98	7·50
Blood volume (litres)	8·59	8·40	8·12	10·58	10·56
Blood pressure	170 / 120	140 / 0	120 / 0	160 / 110	190 / 110

The clinical findings are shown in Table 5. As will be seen, the findings on the two cattle were similar to those on sheep. There were no significant changes in the blood chemistry except a terminal rise in blood sugar and urea nitrogen and a fall in bicarbonate. These were also noted in the sheep.

There was again a fall in the haematocrit. In the case of Bovine No. 1 the marked terminal drop in diastolic arterial blood pressure and rise in venous pressure, were encountered. Both animals died overnight and, in the case of No. 2, the fall in diastolic pressure had not yet occurred at the time of the last examination. In neither case could a final fall in plasma volume be demonstrated.

DISCUSSION

This investigation did not reveal any constant or specific changes in the blood chemistry which could be associated with heartwater.

The main findings were a marked drop in diastolic blood pressure followed by a severe fall in plasma and blood volumes. Both these events took place in the terminal stages of the disease.

The fall in diastolic arterial blood pressure together with a rise in venous pressure can only be ascribed to peripheral vaso-collapse as port mortem examination revealed no abnormalities of the heart valves.

It will be realised that it was extremely difficult to record all phases of this terminal circulatory collapse as it took place very rapidly and almost immediately before death. Furthermore, there was no method of predicting its onset and the majority of deaths took place at night.

That the fall in diastolic pressure could take place very suddenly is well shown in sheep No. 6 (Table 3) where it fell from 85 to 0 between 9 a.m. and 2 p.m.

Peripheral vaso-collapse could be attributed to a sympatholytic effect. That such an effect is present in heartwater is also shown by the haematocrit readings which showed a tendency to fall, often dramatically, during the disease. That this was not due to red cell destruction is suggested by the absence of bilirubinaemia. Turner & Hodgetts (1959) have demonstrated large variations in the haematocrits of sheep due to contraction and relaxation of the spleen and they concluded that the relaxed spleen can contain up to one-seventh of the total blood volume and one-quarter of the total red cell volume. This was confirmed during the present work where it was found that the haematocrit of sheep unaccustomed to being handled was invariably higher than it was when the animals were no longer nervous. The drop in haematocrit values seen in heartwater can, therefore, be ascribed to splenic relaxation following on sympatholysis which culminates in general vaso-collapse.

As would be expected, the haematocrit rose with the sudden reduction of plasma volume, but the extent of this rise was not proportional to the fall in plasma volume. Assuming the total volume of circulating erythrocytes to remain constant, the percentage plasma volume change indicated by a change in the haematocrit can be calculated by the formula—

$$\text{Percentage plasma volume change} = \left\{ \left(\frac{\text{H. crit. 1}}{\text{H. crit. 2}} \right) - 1 \right\} \times 100$$

In all cases the percentage decrease in plasma volume so calculated was considerably less than that shown by direct measurement. This would indicate that a further sludging of red cells takes place in the final stages of the disease. These conclusions are substantiated by the well-known finding of severe engorgement of the spleen at post-mortem examination of animals which have died of heartwater.

A study of the figures from sheep No. 9 and 10 (Tables 2 and 3) and from bovine No. 1 (Table 4) shows that the sudden decrease in plasma volume follows the peripheral vaso-collapse and fall in diastolic pressure. The decrease in plasma volume is not due to hypoproteinaemia as it is not preceded by any reduction in plasma total protein or albumin concentrations. The fact that the plasma protein concentration remains constant despite a sudden reduction in plasma volume indicates that the proteins escape from the vascular system as freely as does the fluid. It is therefore postulated that the decrease in plasma volume is due to a sudden increase in capillary permeability, allowing the escape of plasma proteins. This would naturally lead to severe oedema and shrinkage of plasma volume. This hypothesis is supported by the fact that the transudates in heartwater frequently show coagulation, indicating that even so large a protein as fibrinogen (molecular weight 450,000) has passed through the capillary wall.

No suggestions can be made as to the cause of the nervous symptoms typical of heartwater as shown by the cattle. These symptoms cannot be ascribed to any changes in the blood constituents determined nor to the circulatory collapse which their appearance preceded.

The findings indicate that the main functional disturbance in the terminal stages of heartwater associated with sudden death is a sympatholysis followed by peripheral vaso-collapse, increased capillary permeability and a drastic reduction in blood volume which precipitates general circulatory failure. The indicated supportive

treatment where this series of events appears imminent would be the administration of sympathomimetics and the transfusion of whole blood or preferably packed red cells to maintain blood volume. Adrenaline or noradrenaline should be administered by constant intravenous drip, a procedure usually impracticable in veterinary practice. Therefore, the use of methylamphetamine as a sympathomimetic is suggested. In one of the experimental sheep showing peripheral vaso-collapse, 30 mg. methylamphetamine was injected subcutaneously. Within five minutes the diastolic arterial pressure rose from 0 to 80 mm. Hg. The effect lasted some two hours. Clinically, the indicated dosage and frequency of administration could be judged by the character of the pulse. A bounding " water hammer " type of pulse would indicate decreased peripheral resistance and the need for further treatment. The efficacy of blood or red cell transfusions could best be judged by the haematocrit, which should be kept above normal to supply blood volume. Where a centrifuge is not available, the haemoglobin level could be used as an indicator.

The unexpected finding of the disappearance of the circulating eosinophiles, even before the onset of a febrile reaction, may be of considerable practical importance. When immunizing against heartwater it frequently occurs that individual animals fail to show a febrile reaction. One is therefore left in doubt as to whether they have immunized or not. A second challenge with infected sheep blood into cattle may produce severe and often fatal shock, especially in Jerseys. Where such a challenge is carried out, it often happens that the animals again fail to react and subsequently prove to be immune. If it can be shown that the disappearance of the circulating eosinophiles is a constant phenomenon of the reaction to heartwater, it may well prove a useful technique in determining which animals have immunized. It may also act as an indication of an impending febrile reaction which may require treatment. Further work will be undertaken on these aspects.

A further aspect of the clinical manifestations of heartwater has been studied. It is not uncommon to find that sheep in which the febrile reaction has been controlled by chemotherapy, later pass into a state of collapse and may lie moribund even for days, showing a normal to subnormal body temperature. Such cases invariably end fatally. This usually occurs when treatment has been delayed and nervous symptoms have been evident for some time. The period of survival is naturally prolonged if the animals are housed.

Three such sheep became available for study, which was carried out in collaboration with the Department of Internal Medicine. These animals appeared to be unconscious, the only outward signs of life being slow respiration and intermittent periods during which weak paddling movements were made. Examination proved that the plasma volume was not unduly reduced nor could any abnormalities be found in the blood constituents as determined in our experimental cases. The cerebrospinal fluid was examined and found to be normal.

It would, therefore, appear that in these cases specific therapy had eliminated the infection in time to avert death from circulatory collapse, but that irreversible brain damage had occurred. It can be said that the animals had the appearance of having undergone a functional decerebration. Death would naturally follow from dehydration and inanition. The treatment of such cases would appear to be hopeless.

Summary

The pathological physiology of heartwater has been studied in acute, untreated fatal cases in sheep and cattle. No significant changes could be found in the blood constituents. The main findings were (*a*) disappearance of circulating eosinophiles;

(*b*) sympatholysis manifested firstly by a fall in haematocrit readings due to splenic relaxation followed by a subterminal peripheral vaso-collapse with a dramatic fall in arterial diastolic pressure; and (*c*) a terminal catastrophic drop in plasma volume due to an increased capillary permeability which allows of the passage of plasma proteins from the vascular system.

The use of sympathomimetics and intravenous infusions of packed red cells or whole blood are recommended as supportive treatment in advanced cases. The possible practical applications of the finding of the disappearance of the eosinophiles prior to the onset of the febrile reaction will be investigated further.

Three sheep showing collapse after having received chemotherapy late in the heartwater reaction were studied. These animals were showing general collapse. The condition is described as a functional decerebration with the vegetative functions of the body proceeding more or less normally. The cerebral damage would appear to be irreversible.

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