Experimentally Induced Faciola hepatica Infection in White-tailed Deer I. Clinicopathological and Parasitological Features

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ABSTRACT

Six white-tailed deer (Odocoileus virginianus) and six sheep were inoculated with metacercariae of Fasciola hepatica. Two animals of each species were given 100, 500 or 2500 metacercariae. Clinicopathological features of these infections were determined by analyses of blood samples collected each week from inoculated deer and sheep as well as from two noninoculated animals of each species. One animal in each inoculated group was killed and examined at six weeks postinoculation and the remainder at 15 weeks postinoculation.

Compared with the values obtained from noninoculated controls, eosinophilia, hyperproteinemia and hyperglobulinemia occurred in inoculated deer. There were no other significant changes in hematological values or in serum aspartate aminotransferase levels. Marked leukocytosis and eosinophilia, with hyperproteinemia, hyperglobulinemia, hypoalbuminemia, elevated serum aspartate aminotransferase levels and mild macrocytic normochromic anemia characterized the infection in lambs.

Although approximately 29% of the inoculum was recovered from the hepatic parenchyma of the sheep, F. hepatica was found in only one of six inoculated deer. A patent in-

Submitted July 16, 1974.

Vol. 39 — April, 1975

fection was established in this deer and constitutes the second report of mature F. hepatica in this host.

RÉSUMÉ

On a administré des métacercaires de Fasciola hepatica à six cerfs de Virginie (Odocoileus virginianus) et à six agneaux. Deux sujets de chacune de ces deux espèces reçurent respectivement: 100, 500 et 2500 métacercaires. On détermina les caractéristiques clinico-pathologiques de ces parasitoses par l'analyse d'échantillons de sang hebdomadaires des cerfs de Virginie et des agneaux expérimentaux, ainsi que de deux sujets témoins de chacune de ces deux espèces. On procéda à l'euthanasie et à la nécropsie d'un sujet de chacun des groupes expérimentaux, au bout de six semaines; on fit la même chose avec le restant des sujets, neuf semaines plus tard.

Comparativement aux témoins, les cerfs de Virginie expérimentaux développèrent une éosinophilie, une hyperprotéinémie et une hyperglobulinémie. On ne décela pas d'autre changement appréciable dans leur formule sanguine ou dans la teneur de leur sérum en transaminase aspartique-cétoglutarique. Chez les agneaux, la distomiase produisit une leucocytose et une éosinophilie élevées, ainsi qu'une hyperprotéinémie, une hyperglobulinémie, une hypoalbuminémie, une augmentation de la teneur du sérum en transaminase aspartiquecétoglutarique et une légère anémie macrocytaire et normochrome.

En dépit du fait qu'on recouvra environ 29% de l'inoculum dans le parenchyme hépatique des agneaux, on ne décela la présence de F.

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This work was supported by the Ontario Ministry of Agriculture and Food, Grant No. 459 from the Canadian National Sportsmen's Show, and the National Research Council of Canada which awarded a Post-Doctoral Fellowship to the senior author.

hepatica que chez un des six cerfs de Virginie. Il se produisit une distomiase évidente chez cet animal, ce qui constitue le second rapport relatif à la présence de F. hepatica adultes chez le cerf de Virginie.

INTRODUCTION

The common liver fluke Fasciola hepatica has been reported from black-tailed deer (Odocoileus hemiorus columbianus) in California (4, 11, 30) and red deer (Cervus elaphus) (1, 17), roe deer (Capreolus capreolus) (1, 17, 33) and elk (Alces alces) (38) in Europe. However, among these hosts the range of susceptibility to infection is apparently wide. Recent experimental work by Barth and Schaich (2) has demonstrated that F. hepatica is more pathogenic in roe deer than in red deer. Kistner and Koller (15) have shown that blacktailed deer can be readily infected, acquiring acute fascioliasis, whereas we have found that white-tailed deer were markedly resistant (21). In the latter host very little migration occurred in the liver and the flukes were rapidly destroyed (21).

As a result of this observation, a second experiment was conducted to evaluate the effects in white-tailed deer of various dose levels of F. hepatica metacercariae over a prolonged period of time. For comparison, sheep were given similar inoculations to induce subacute, chronic and subclinical fascioliasis (3, 28). The present paper records the clinicopathological and parasitological features of these infections.

MATERIALS AND METHODS

INOCULUM

Metacercariae of F. hepatica were collected from laboratory reared Lymnaea columella. These snails were artificially exposed to miracidia hatched from fluke eggs obtained from infected sheep in Oregon.

DEER

The deer used were part of a captive herd maintained in a 0.37 hectare enclosure at the Ontario Veterinary College. Before the experiment began eight fawns, five to six months of age, were immobilized with a combination of etorphine hydrochloride and xylazine (22) and transported to a modified boxstall where they could be physically restrained for collection of blood samples. They were given levamisole hydrochloride¹ as a drench at the dose rate of 16 mg/kg of body weight to reduce existing gastrointestinal nematode burdens.

On the day of inoculation the fawns were immobilized, weighed and metacercariae were administered orally by means of a stomach tube. Two fawns (No. 1 and 2) were noninoculated controls (group I), two (No. 3 and 4) were given 100 metacercariae (group II), two (No. 5 and 6) 500 metacercariae (group III) and two (No. 7 and 8) 2,500 metacercariae (group IV). One fawn in each inoculated group was killed and examined at approximately 42 days PI and remaining fawns on days 105 and 106.

During the experiment, one noninoculated control (No. 2) died with signs of acute hemorrhagic enteritis on day 52 postinoculation (PI). Blood was passed with feces during the 12 hour period before it died. Escherichia coli was cultured from the liver. lung, spleen, lymph nodes and intestines of this fawn. Fawn No. 4 (100 metacercariae) died with similar signs on day 62 PI. In addition to E. coli, Clostridium sp. was isolated from the liver, lung and intestines but toxin was not demonstrated by mouse inoculation. Surviving fawns were injected intramuscularly with choloramphenicol² for five consecutive days (63 to 67 PI) and this antibiotic³ was added to the drinking water for a three week period beginning on day 63 PI.

Sheep

Eight six month old crossbred (Cheviot X Lester) lambs were the host controls for the inoculations. They were given levamisole, weighed, grouped and inoculated in a manner similar to that used for the fawns.

¹Tramisol, Cyanamid of Canada Limited, Agricultural Products Department, Montreal, Quebec.

²C-150 Chloramphenicol Injectable, W. E. Saunders, Ltd., London, Ontario.

³Anivon, Rogar/STB, Division of BTI Products Inc., St. Hyacinthe, Quebec.

BLOOD ANALYSES

Blood samples were collected at weekly intervals from the fawns and lambs. Several hematological and serum biochemical analyses were made. Methods used for blood collection and analyses of the various parameters were given previously (20). Additional tests were conducted on serum samples collected from the fawns on days 35 and 105 PI and the lambs on days 42 and 105. Commercial kits and their specified methods were used to determine levels of serum alanine aminotransferase (GPT)⁴. alkaline phosphatase (AP)⁵ activity and bilirubin concentration⁶. Serum GPT levels were determined at 30°C and AP at 37°C and values were expressed in international units (IU).

Hematological and serum biochemical data were not statistically analysed because of the small number of animals in each group. The mean values and standard errors for each parameter were calculated for samples collected from fawns and lambs prior to inoculation. Indication of possible significant change in blood values was obtained by comparing the values obtained during the experiment with those from preinoculation samples and with the controls.

FECAL EXAMINATION

Fecal samples, collected from the fawns and lambs before the experiment began and on several occasions during the experiment, were examined for all types of gastrointestinal nematode eggs. Quantative total eggs per gram of feces (EPG) counts were determined by the Cornell-McMaster method (10). Fecal examination for F. hepatica eggs was done on samples collected from the lambs each week beginning on day 63 PI. For the fawns it was done at least once every two weeks. Quantitative fluke EPG counts were determined by the method described by Knapp and Presidente (16).

NECROSPY PROCEDURE

The methods used at the time of necropsy were previously given (21). Contents of the abomasum and small and large intestines were collected over a 100 mesh sieve from five inoculated fawns. The numbers of nematodes recovered in 10% of the content from the abomasum and small intestine were used to estimate the total worm burden in these organs. All content from the large intestine was examined for nematodes. The worms were fixed in 10%formalin and later identified.

RESULTS

CLINICAL SIGNS

Fawns — Clinical signs associated with F. hepatica infection were not seen in inoculated fawns. These fawns, with the possible exception of those in group IV, gained as much weight as did noninoculated controls (Table I).

Lambs — Distention of the abdomen and a slight paleness of mucous membranes of lamb No. 8 were the only signs evident among inoculated lambs. Lambs given F. *hepatica* did not gain as much weight as the controls (Table I).

CLINICOPATHOLOGICAL FEATURES

The mean preinoculation values for the hematological and serum biochemical parameters examined in the fawns and lambs are given in Table II. Values obtained from the noninoculated controls during the experiment were within the range of values established from the preinoculation sample.

HEMATOLOGICAL DATA

Fawns — Notable changes in these parameters did not occur after inoculation, except in one fawn. In fawn No. 8 hemoglobin concentration (Hb) was reduced (approximately 15.8 g/100 ml) and the erythrocyte count (RBC) decrease (approximately 13.1 x 10⁶/mm³) on days 56, 63 and 70 PI. The mean corpuscular volume (MCV, $42.5\mu^3$) and mean corpuscular hemoglobin (MCH, 15.8 pg) were increased on day 98 PI. Reticulocytes were not seen in any of the blood smears although some macrocytic erythrocytes were found in smears from fawn No. 8. Prevalence of sickled cells was

^{*}SGPT kit, Calbiochem, San Diego, California.

⁵Phosphatrate Alkaline klt, Warner Chilcott, Morris Plains, New Jersey.

American Monitor Corporation, Indianapolis, Indiana.

greater in smears stained in new methylene blue two to four hours after collection than was found in smears from the same fawns made at the time of sample collection. A marked increase in the total leukocyte count (WBC) for each surviving fawn was found on day 70 PI (Fig. 1). This was due to an increase in circulating neutro-

TABLE I. Initial Body Weights (kg) and Change in Weight (kg) for Fawns and Lambs Given Me-
acercariae (MC) of Fasciola hepatica

			Fawns		Lambs			
Group	Animal No.	On Day 0	Weight Change	No. of Days	On Day 0	Weight Change	No. of Days	
I	$\frac{1}{2}$	25 34	+5 +3	105 52	24 31	$^{+4}_{+4}$	1C5 105	
II (100 MC)	3 4	25 34	$^{+5}_{+2}$	43 62	24 30	$^{0}_{+3}$	42 105	
III (500 MC)	5 6	34 31	$^{+4}_{+3}$	44 105	30 24	$^{+2}_{0}$	43 106	
IV (2500 MC)	7 8	35 24	$^{+2}_{+2}$	46 106	26 28	$^{+1}_{0}$	43 107	

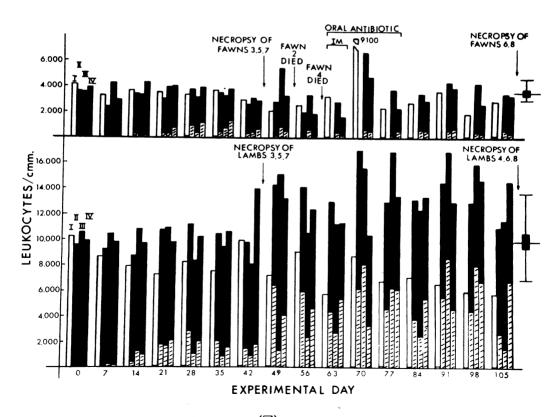


Fig. 1. Total leukocyte and absolute cosinophil counts (\square) for Fasciola hepatica — inoculated fawns and lambs (\blacksquare) given 100 (group II), 500 (group III) and 2500 (group IV) metacercariae and for noninoculated fawns and lambs (\square) group I). Mean total leukocyte counts for samples collected prior to inoculation are 3790 \pm 220/mm³ for these eight fawns and 9910 \pm 520/mm³ for these eight lambs.

phils. After day 77 PI the WBC count for inoculated fawns was slightly increased over that of fawn No. 1. The mean absolute eosinophil count for group I fawns was $87/\text{mm}^3$ (minimum and maximum, 22 and 144) during the experiment. The eosinophil counts for inoculated fawns increased on day 14 PI and remained elevated until the time of necropsy (Fig. 1). The highest eosinophil count (45% of the total leukocytes) was found in fawn No. 8 on day 35 PI.

Lambs — Changes in hematological values were seen towards the end of the experiment, particularly in lambs given 500 and 2500 metacercariae (No. 6 and 8). In these lambs, decreases in Hb (to 8.6 g/100 ml) and RBC (to 7.24 x 10^6 /mm³) began on day 84 PI. The PCV was 20% for lamb No. 8 on day 105 PI. Reticulocytes were not seen in blood smears from the inoculated lambs although macrocytic erythrocytes were commonly found. The WBC counts for inoculated lambs were elevated on day 49 PI and remained so until the end of the experiment (Fig. 1). The mean absolute eosinophil count for control lambs was $87/\text{mm}^3$ (minimum and maximum, 31 and 150) during the experiment. In inoculated lambs, eosinophil counts began to increase on day 14 PI and an early peak (23% of WBC) was evident in group II lambs on day 28. Peak eosinophil counts in lambs after day 42 PI occurred on days 70 (No. 4, 36% of WBC), 91 (No. 6, 51% of WBC) and 98 (No. 8, 46% of WBC) (Fig. 1).

SERUM PROTEIN CONCENTRATION

Fawns — Increased total protein (TP) levels were found in all inoculated fawns on days 28, 35 and 42 PI. This was most pronounced in fawns in group IV on day 35 PI when TP was increased an average of 1.9 g/100 ml over their preinoculation values. This elevation was due to increases in

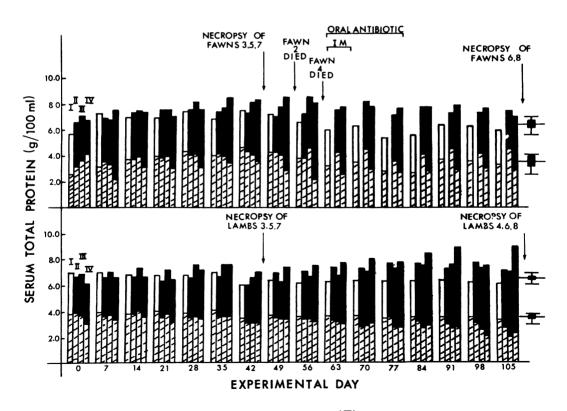


Fig. 2. Serum total protein (TP) and albumin (SA) concentrations (7) for Fasciola hepatica — inoculated fawns and lambs (\blacksquare) given 100 (group II), 500 (group III) and 2500 (group IV) metacercariae and for noninoculated fawns and lambs (\square group I). Mean values for sample collected prior to inoculation are: TP, 6.5 \pm 0.3 and SA, 3.6 \pm 0.4 g/100 ml for four of these fawns and TP, 6.6 \pm 0.3 and SA, 3.6 \pm 0.4 g/100 ml for seven of these lambs.

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all globulin fractions, especially the β group. This increase was not as evident when these values were compared with the mean value for the noninoculated controls during this time (Fig. 2). This was because one of the controls had higher TP levels (by 1.5-2.8 g/100 ml) than the preinoculation mean value for all fawns (Table II). After day 42, TP levels gradually declined in all inoculated fawns. Consistent changes in serum albumin concentration (SA) were not evident during the experiment.

Lambs — On day 35 PI the earliest rise in TP occurred. Levels for lambs in group IV were an average of 1.2 g/100 ml greater than their preinoculation values. After this time TP increased gradually in all inoculated lambs. Peak levels were attained after day 70 PI (Fig. 2) and this increase was due primarily to a rise in γ globulins. In inoculated lambs SA began to decrease on day 63 PI and was lowest on days 98 and 105.

SERUM BIOCHEMICAL ANALYSES

Fawns — Serum aspartate aminotransferase (GOT) levels were elevated in early blood samples collected from the fawns. The mean value on day 21 was taken as "normal" for these fawns (Table II). A marked increase in GOT for all fawns was seen only on day 65 PI (Fig. 3).

Significant changes in GPT, AP, bilirubin concentration, serum iron concentration (SI) and total iron binding capacity (TIBC) were not found in samples collected from inoculated fawns on day 35 and 105 PI.

Lambs — In inoculated lambs, increases in GOT began on day 21 PI, peaked on days 63 and 70 and then gradually declined (Fig. 3).

There were no significant changes in SI and TIBC in inoculated lambs on day 35 PI. However, on day 105, SI was markedly reduced in lamb No. 6 (35 mg/100 ml com-

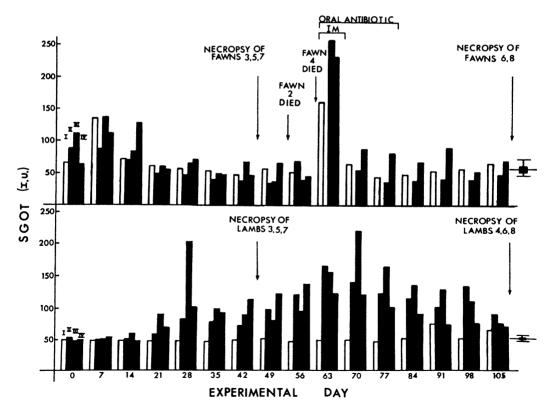


Fig. 3. Serum aspartate aminotransferase (GOT) values for Fasciola hepatica-inoculated fawns and lambs (\blacksquare) given 100 (group II), 500 (group III) and 2500 (group IV) metacercariae and for noninoculated fawns and lambs (\square group I). Mean values for samples collected prior to inoculation are: 56 \pm 3.7 international units (IU) for these eight fawns on day 21 and 51 \pm 1.3 IU for seven of these lambs on day 0.

	Fa	wns	Lambs			
Itemª	Mean Value and Standard Error	Minimum and Maximum	Mean Value and Standard Error	Minimum and Maximum		
Hb, g/100 ml. PCV, %	$\begin{array}{c} 17.9 \ \pm \ 0.6 \\ 48 \ \pm \ 1.1 \\ 17.5 \ \pm \ 0.6 \\ 27 \ \pm \ 0.7 \\ 10.3 \ \pm \ 0.2 \\ 37.6 \ \pm \ 0.4 \\ 3790 \ \pm \ 220 \\ 6.5 \ \pm \ 0.3 \\ 2.9 \ \pm \ 0.4 \\ 3.6 \ \pm \ 0.4 \\ 56 \ \pm \ 3.7^{\mathrm{b}} \end{array}$	$\begin{array}{rrrrrrrrrrrrrrrrrrrrrrrrrrrrrrrrrrrr$	$\begin{array}{c} 12.7 \ \pm \ 0.4 \\ 35 \ \pm \ 0.9 \\ 12.1 \ \pm \ 0.3 \\ 29 \ \pm \ 0.6 \\ 10.4 \ \pm \ 0.2 \\ 36.5 \ \pm \ 0.6 \\ 9910 \ \pm \ 520 \\ 6.6 \ \pm \ 0.3 \\ 3.0 \ \pm \ 0.2 \\ 3.6 \ \pm \ 0.4 \\ 51 \ \pm \ 1.3 \end{array}$	$\begin{array}{rrrrrrrrrrrrrrrrrrrrrrrrrrrrrrrrrrrr$		

TABLE II. Mean Hematological and Serum Biochemical Values for Fawns and Lambs Collected Before Time of Inoculation with Metacercariae of Fasciola hepatica

Abbreviations are: Hb, hemoglobin concentration; PCV, packed cell volume; RBC, erythrocytes; MCV, mean corpuscular volume; MCH, mean corpuscular hemoglobin; MCHC, mean corpuscular hemoglobin concentration; WBC, leukocytes; TP, total protein concentration; SA, serum albumin concentration; and GOT, serum aspartate aminotransferase activity measured in international units •Value determined from samples collected on day 21 postinoculation

pared to 111 mg/100 ml for controls) and TIBC was elevated in inoculated lambs (429 to 621 mg/100 ml compared to 359 mg/100 ml for controls). Bilirubin concentration and GPT levels remained within normal range in inoculated lambs and no consistent change was found in AP activity.

PARASITOLOGICAL FINDINGS - FECAL SAMPLE EXAMINATION

Fawns - Total nematode EPG counts on experimental day 0 indicated that treatment with levamisole did not remove entirely the existing nematode burdens (Table III). Eggs of F. hepatica were found only in feces from fawn No. 8. They were first recovered in a sample on day 84 PI and the fawn was passing 24 EPG on day 105. The mean size of 20 fluke eggs was 124 μ (115-130) X 67µ (62-72).

Lambs — Treatment with levamisole reduced the high total nematode EPG counts found in the lambs before the experiment began (Table III). Fluke eggs were first recovered in feces from lamb No. 4 on day 77 PI, then in lamb No. 6 on day 91 and in lamb No. 8 on day 105. The mean size of 20 fluke eggs was 126 μ (115-139) X 67 μ (58-77).

RECOVERY OF FLUKES

Fawns — Immature F. hepatica was not

recovered from the livers of fawns examined on days 43, 44, 46 or 62 PI (Table IV) but lesions associated with migrating flukes were found (described in part II). Flukes were not found in fawn No. 6 but 32 F. hepatica were recovered from fawn No. 8 (Fig. 4). The mean length of the flukes was 22 mm and most were mature (Table IV). Some of these specimens have

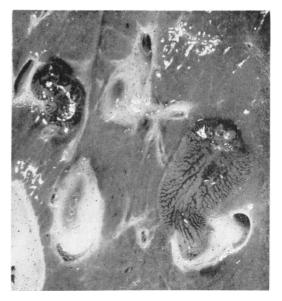


Fig. 4. Mature Fasciola hepatica in bile ducts seen on cut surface of the liver from fawn No. 8. This fawn was given 2500 metacercariae of F. hepatica and examined on day 106 postinoculation.

TABLE III. Total Fecal Egg per Gram (EPG) Counts for Fawns and Lambs given Metacercariae
(MC) of Fasciola hepatica. Nematode EPG Counts before Levamisole Treatment (Pre-trt) on Ex-
perimental day 0 and at Time of Necropsy and Fluke EPG Counts at Time of Necropsy

		Fawns				Lambs			
Group	Animal No.	Nematode EPG Counts				Nematode EPG Counts			
		Pre- trt ^a	On day 0	At Necropsy	Fluke EPG	Pre- trt	On day 0	At Necropsy	Fluke ¹ EPG
I	$\frac{1}{2}$	_	50 50	50	0 0	900 1050	0 0	50 50	0
II	3 4		100 200	50	0 0	50 2500	0 0	0 0	0 42
III	5 6	_	500 150	650 150	0 0	2350 200	0 0	0 0	0 1
IV (2500 MC)	7 8	_	250 750	0 150	0 24	600 7250	0 0	0 0	0 <1

^aQuantitative flotation examination of fecal samples from several of these fawns indicated moderate to large numbers of gastrointestinal nematode eggs and necessity for anthelmintic therapy

TABLE IV. Recovery of *Fasciola hepatica* from Fawns and Lambs at the Time of Necropsy: Number and Length (mm) of Flukes Recovered

Group		F. hepatica from Fawns				F. hepatica from Lambs			
	Animal No.			finmax. length	No. meas.	No. of flukes		Minmax. length	No. meas.
I	$\frac{1}{2}$	0	_						_
II	3 4	0 0				33 50	7 22	$\begin{array}{ccc} 4 & - & 9 \\ 9 & - & 31 \end{array}$	16 38
III	5 6	0 0	_	_	_	163 138	5 16	3 - 7 4 - 26	25 100
IV (2500 MC)	7 8	0 32	22	13 — 28	23	715 150	4 10	$2 - 7 \\ 4 - 24$	35 94

been deposited in the National Parasite Collection and accessioned as U.S. National Helminth Collection No. 72743.

Approximately 280 Ostertagia (minimum and maximum, 0 and 870) were recovered from five inoculated fawns. Burdens of 300 Nematodirus were found in fawn No. 3 and 5. Small numbers of Capillaria, Cooperia, Oesophagostomum and Trichuris were also recovered from some of these fawns.

Lambs — The numbers and lengths of the flukes recovered from inoculated lambs are summarized in Table IV. Approximately 29% of the inoculum was recovered from the hepatic parenchyma (6 weeks PI). Flukes recovered from lambs given 500 and 2500 metacercariae were smaller than those from lambs given 100 metacercariae (Table IV). The fluke burden (150) for lamb No. 8 examined on day 105 PI was greatly reduced when compared to that found in lamb No. 7 (715) on day 43.

DISCUSSION

Hematological values obtained from the fawns before inoculation and from the noninoculated controls (group I) during the experiment agree with published data for white-tailed deer of similar age (6, 14, 31, 34, 39). The increase in leukocyte counts for all fawns on day 65 PI (Fig. 3) was attributed to an increase in neutrophils following infection that resulted in the death of two fawns. Absolute eosinophil counts for group I fawns were low and the percent of total leukocytes (1-5%) was similar to

that reported by Teeri et al (31) (0-5%)but less than the 18% given for one year old deer by Dommert et al (6). Serum protein concentrations were similar to or lower than those recorded by others (14, 34, 39). Serum biochemical data also compared favorably with values given for white-tailed deer (26, 34, 35, 36). The increase in GOT values for all fawns on day 65 PI was associated with leakage of the enzyme from muscle injured during physical restraint and intramuscular injection of antibiotics. This blood sample was collected on the third day that the fawns had been restrained to inject chloramphenicol. This response is consistent with data reported by Seal et al (26) who demonstrated a marked increase in levels of GOT and creatine phosphokinase activity 24 hours after deer had been physically restrained.

In inoculated fawns, eosinophilia, hyperproteinemia and hyperglobulinemia were the major changes that occurred in hematological and serum biochemical values. Changes in other hematological parameters, SA concentration or GOT activity were not found. The presence of small gastrointestinal nematode burdens in inoculated fawns evidently had little effect on the blood parameters analyzed.

Preinoculation and control blood values of lambs were within the normal limits given for sheep (12). In lambs that were inoculated with F. hepatica a mild normochromic anemia began during the last three weeks of the experiment. Marked eosinophilia with leukocytosis and serum hyperproteinemia, hypergammaglobulinemia with hypoalbuminemia were found. These changes are characteristic for sheep infected with F. hepatica (3, 12, 24, 25, 27, 28, 29). Elevated GOT levels in the inoculated lambs were consistent with data given for F. hepatica infected sheep (12, 24, 25, 32). Decrease in SI concentration in one lamb (No. 6) has also been reported by previous workers (28).

Fasciola hepatica was not recovered from five of six inoculated fawns and this is similar to observations made on three deer examined at earlier stages of infection (21). These data provide additional evidence to show that white-tailed deer are strongly resistant to F. hepatica. Mature F. hepatica was recovered from one fawn (No. 8) but only a small percent (1.3%) of the inoculum was recovered. The prepatent period and size of flukes in this fawn were similar to

those recovered from lamb No. 4 (Table IV). To our knowledge this is the first published report of a patent infection in white-tailed deer. Naturally occurring infections have not been found in areas where deer share range with infected cattle (23) and F. hepatica is not listed as a parasite of white-tailed deer (37). Recently, Foreyt (8) informed us that patent F. hepatica infection was induced in three white-tailed deer. A single mature F. hepatica was recovered from each of two deer inoculated orally with small numbers of metacercariae (5 and 50). A mature fluke was also recovered from a deer inoculated intraperitoneally with six metacercariae (8).

The mean percent (29%) of the inoculum that was recovered from the livers of the lambs at six weeks PI corresponds closely to the results of others (3, 28). Fluke EPG counts and size of F. hepatica recovered from inoculated lambs indicated a retarded maturation rate in lambs given 500 or 2500 metacercariat. This response is known to occur in many parasitic infections and is typical for F. hepatica in sheep (3, 24, 25, 28). The response of F. hepatica infected lambs to the three dose levels confirms the infectivity of the metacercariae used in this experiment and shows clearly the marked resistance of white-tailed deer to this liver fluke.

The reaction of swine which are known to be resistant to F. hepatica (5, 13, 18) is similar to that of white-tailed deer. In both hosts the infection is characterized by a low recovery of inoculum and a similar response in circulating eosinophils. In swine, F. hepatica was rapidly walled off and destroyed and only small numbers matured (5, 7, 13, 18). Enigk and Dey-Hazra (7) showed that in swine given 500 or 800 metacercariae, GOT, GPT and γ -glutamyltranspeptidase levels were moderately increased between four and seven weeks. PI.

Recently, red and roe deer were experimentally infected with sheep or bovine isolates of F. hepatica (2). These Eurasian cervids are both susceptible to F. hepatica and host response to infection was different from that seen in white-tailed deer. The prepatent period for bovine strain F. hepatica in roe deer was 49 to 51 days and for sheep strain 52 to 59 days. In red deer it was slightly longer (2). This is shorter by approximately four weeks than the prepatent period for F. hepatica in white-

tailed deer (76 to 84 days (8), the present experiment). The mean size of eggs for sheep strain F. hepatica in roe deer was the same as that in white-tailed deer in the present experiment. Barth and Schaich (2) found at 14 weeks PI that an average of 10.3% of sheep strain flukes were recovered in roe deer (28.2% of this strain were recovered from red deer but 31% of these flukes were immature). This is greater than the percentage recovery (0 and 1.3%) found in the two white-tailed deer examined at this time in the present experiment. The mean size of flukes recovered in roe deer was the same as that for flukes recovered in the one white-tailed deer with a patent infection. Clinical signs and significant clinicopathological changes were not seen in infected roe and red deer (2). The increase in eosinophil counts in roe deer was not significant (2) whereas in white-tailed deer it was marked. Leukocytosis was not a characteristic feature of F. hepatica infection in any of these cervid hosts.

In six black-tai'ed deer given 250, 500 or 1000 metacercariae the mean percentage recovery of these doses was 37% (minimum and maximum 24 and 54) (15). This was less than the mean recovery (51%, minimum and maximum 44 and 72) from three sheep given the same doses. However, the size of the flukes recovered from the blacktailed deer was greater than from the sheep and the legions associated with F. hepatica infection were more severe in the deer than in the sheep. Also, these data provide additional confirmation for the infectivity of the metacercariae used in our experiment since the metacercariae used in both the black-tailed and white-tailed deer experiments were part of the same batch (15).

ACKNGWLEDGMENTS

The authors wish to thank Dr. T. P. Kistner, Department of Veterinary Medicine, Oregon State University for providing the metacercariae used in this investigation. We gratefully acknowledge the technical assistance of personnel in the Clinical Pathology section and Mr. Ted Eaton, Department of Pathology and Mr. Hugh Belcher, animal technician, Edinburgh Road Research Station, Ontario Veterinary College.

REFERENCES

- 1. BARTH, D. and K. SCHAICH. [The occurrence of Fasciola hepatica in roes (Capreolus capreolus) and red deer (Cervus elaphus) and its control with rafoxanide.] Dt. tierärztl. Wschr. 80: 420-424; 448-450. 1978.
- BARTH, D. and K. SCHAICH. [Investigations on the experimental fasciolosis in roes (Capreolus capreolus) and red deer (Cervus elaphus).]Z. Jagdwiss. 19: 183-197. 1973.
- BORAY, J. C. Studies on experimental infections with Fasciola hepatica, with particular reference to acute fascioliasis in sheep. Ann. trop. Med. Parasit. 61: 439-450. 1967.
- 4. BROWNING, B. M. and E. M. LAUPPE. A deer study in a redwood-Douglas fir forest type. Calif. Fish and Game 50: 132-137. 1964.
- DALCHOW, W. and F. HÖRCHNER. [Experimental infection with Fasciola hepatica in different animals.] Berl. Münch. tierärztl. Wschr. 85: 271-274. 1972.
- 6. DOMMERT, A. R., M. E. TUMBLESON, R. B. WESCOTT, D. A. MURPHY and L. J. KORSCHGEN. Hematologic values for dieldrin-treated white-tailed deer (Odocoilens virginianus) in Missouri. Am. J. vet. clin. Path. 2: 181-184. 1968.
- ENIGK, K. and A. DEY-HAZRA. [Mineral content and enzyme activity of the blood plasma and mineral content of the liver in the pig during prepatent fasciolosis.] Dt. tierärztl. Wschr. 80: 541-564. 1973.
- FOREYT, W. J. Personal communication. Department of Veterinary Science. University of Wisconsin, Madison, Wisconsin. 1973.
- 9. FOREYT. W. J. and A. C. TODD. The occurrence of Fascioloides magna and Fasciola hepatica together in the livers of naturally infected cattle in south Texas and the incidence of the flukes in cattle, white-tailed deer and feral hogs. J. Parasit. 58: 1010-1011. 1972.
- GEORGI, J. R. Quantitative fecal examination. In Parasitology for Veterinarians. pp. 173-175. Toronto: W. B. Saunders Co. 1974.
- 11. HFRMAN, C. M. Some worm parasites of deer in California. Calif. Fish and Game 31: 201-208. 1945.
- HJERPE. C. A., B. C. TENNANT, G. L. CRENSHAW and N. F. BAKER. Ovine fascioliasis in California. J. Am. vet. med. Ass. 159: 1266-1271. 1971.
- HÖRCHNER, F. and W. DALCHOW. [Experimental Fasciola hevatica infection in pigs.] Berl. Münch. tierärztl. Wschr. 85: 184-188. 1972.
- 14. JOHNSON, H. E., W. G. YOUATT, L. D. FAY, H. D. HARTE and D. E. ULLREY. Hematolovic values of Michigan white-tailed deer. J. Mammal. 49: 749-754. 1968.
- KISTNER, T. P. and L. D. KOLLER. Experimentally induced Fasciola hepatica infections in black-tailed deer. J. Wildl. Dis. 11: 214-220. 1975.
- KNAPP, S. E. and P. J. A. PRESIDENTE. Efficacy of rafoxanide against natural Fasciola hepatica infections in cattle. Am. J. vet. Res. 32: 1289-1291.
- KOTRLÝ, A. [The mutual relation of the parasites of ungulate game and domestic animals.] Vet. Med. Praha 12: 745-752. 1967.
- NANSEN, P., S. ANDERSEN, E. HAPMER and H.-J. RIISING. Experimental fascioliasis in the pig. Expl Parasit. 31: 247-254. 1972.
- PRESIDENTE, P. J. A., S. E. KNAPP and K. D. NICOL. Pathogenicity of experimentally induced concurrent infections of Fasciola henatica and Haemonchus contortus in sheep. Am. J. vet. Res. 34: 51-60. 1973.
- 20. PRESIDENTE, P. J. A., J. H. LUMSDEN, K. R. PRESNELL, W. A. RAPLEY and B. M. McCRAW. Combination of etorphine and xylazine in captive white-tailed deer: II. Effects on hematologic, serum biochemical and blood gas values. J. Wildl. Dis. 9: 342-348. 1973.
- 21. PRESIDENTE, P. J. A., B. M. McCRAW and J. H. LUMSDEN. Early pathological changes associated with Fasciola hevatica infection in white-tailed deer. Can. J. comp. Med. 38: 271-279. 1974.

- 22. PRESNELL, K. R., P. J. A. PRESIDENTE and W. A. RAPLEY. Combination of etorphine and xylazine in captive white-tailed deer: I. Sedative and immobilization properties. J. Wildl. Dis. 9: 336-341, 1973.
- PRESTWOOD, A. K. and F. E. KELLOGG. Helminth parasitism among intermingling insular populations of white-tailed deer, feral cattle and feral swine. Program and Abstracts of the American Society of Parasitologists. 48th Annual Meeting. p. 38. 1973.
- 24. PULLAN, N. B., M. M. H. SEWELL and J. A. HAMMOND. Studies on the pathogenicity of massive infections of Fasciola hepatica L. in lambs. Br. vet. J. 126: 543-557. 1970.
- ROBERTS, H. E. Observations on experimental acute fascioliasis in sheep. Br. vet. J. 124: 433-450. 1968.
- SEAL, U. S., J. J. OZOGA, A. W. ERICKSON and L. J. VERME. Effects of immobilization on blood analyses of white-tailed deer. J. Wildl. Mgmt. 36: 1034-1040. 1972.
- SEWELL, M. M. H., J. A. HAMMOND and D. C. DINNING. Studies on the aetiology of anaemia in chronic fascioliasis in sheep. Br. vet. J. 124: 160-170. 1968.
- SINCLAIR, K. B. Pathogenesis of Fasciola and other liver-flukes. Helminth. Abstr. 36: 115-134. 1967.
- SYMONS, L. E. A. and J. C. BORAY. The anaemia of acute and chronic ovine fascioliasis. Z. Tropenmed. Parasit. 19: 451-472. 1968.
- TABER, R. D. and R. F. DASMANN. The blacktailed deer of the chaparral. Its life history and management in the north coast range of California.

Game Bull. 8. Calif. Dept. Fish and Game. 163 pp. 1958.

- TERRI, A. E., W. VIRCHOW, N. G. COLOVOS and F. GREELEY. Blood composition of whitetailed deer. J. Mammal. 39: 269-274. 1958.
- THORPE, E. and E. J. H. FORD. Serum enzyme and hepatic changes in sheep infested with Fasciola hepatica. J. Path. 97: 619-629. 1969.
- TOMÁNEK, J. [On the identification of the helminthofauna of roe-deer in Northern Moravia.] Vet. Med. Praha 12: 739-744. 1967.
- 34. TUMBLESON, M. E., J. D. CUNEIO and D. A. MURPHY. Serum biochemical and hematological parameters of captive white-tailed fawns. Can. J. comp. Med. 34: 66-71. 1970.
- 35. TUMBLESON, M. E., J. W. TICER, A. R. DOM-MERT, D. A. MURPHY and L. J. KORSCHGEN, Serum proteins in white-tailed deer in Missouri. Am. J. vet. clin. Path. 2: 127-131. 1968.
- 36. TUMBLESON, M. E., M. G. WOOD, A. R. DOM-MERT, D. A. MURPHY and L. J. KORSCHGEN. Biochemic studies on serum from white-tailed deer in Missouri. Am. J. vet. clin. Path. 2: 121-125. 1968.
- 37. WALKER, M. L. and W. W. BECKLUND. Checklist of the internal and external parasites of deer, Odocoileus hemionus and O. virginianus, in the United States and Canada. Index-Cat. med. vet. Zool., Special Publ. No. 1, 45 pp. 1970.
- WETZEL, R. and K. ENIGK. Zur wurmfauna des elches. Dt. tierärztl. Wschr. 44: 576-577. 1936.
- WHITE, M. and R. S. COOK. Blood characteristics of free-ranging white-tailed deer in southern Texas. J. Wildl. Dis. 10: 18-24. 1974.